JOURNAL OF AGRICULTURAL RESEARCH

VOLUME 53

JULY 1–DECEMBER 15, 1936

ISSUED BY AUTHORITY OF THE SECRETARY OF AGRICULTURE
WITH THE COOPERATION OF THE ASSOCIATION OF
LAND-GRANT COLLEGES AND UNIVERSITIES

UNITED STATES
GOVERNMENT PRINTING OFFICE
WASHINGTON : 1937

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Published on the 1st and 15th of each month. This volume will consist of 12 numbers and the contents and index.

Subscription price:
Entire Journal: Domestic, $3.25 a year (2 volumes).
Foreign, $4.75 a year (2 volumes).
Single numbers: Domestic, 15 cents.
Foreign, 20 cents.

Articles appearing in the Journal are printed separately and can be obtained by purchase at 5 cents a copy domestic; 8 cents foreign. If separates are desired in quantity, they should be ordered at the time the manuscript is sent to the Printer. Address all correspondence regarding subscriptions and purchase of numbers and separates to the Superintendent of Documents, Government Printing Office, Washington, D. C.

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JOURNAL OF AGRICULTURAL RESEARCH

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ERRATA AND AUTHORS' EMENDATIONS

Page 3, line 16, add "25° C."

Page 9, line 2, "either sucrose or reducing sugars" should be "sucrose content."

Page 76, table 3, Soil no. 312, Mar. 2, Eh "+.030" should be "−.030."

Page 102, table 4, last line of heading, "July bale..ce" should be "Joly balance."

Page 110, table 6, in third line of heading, "0" should be "0.5."

Page 153, line 7, "earliest culm" should be "tallest culm."

Page 155, sixth line from bottom, "greater portion" should be "a large number."

Page 189, table 2, the first brace is incorrectly shortened by one number, thereby making all subsequent brace inclusions wrong except the last.

Page 229, table 2. The following note should be inserted immediately above the footnotes: "In 1932, normal cows 2, 3, 4, and 6 gave negative reactions; in 1933, udderless-control cow 35 and normal-control cows 6 and 7 were negative; in 1934, udderless-control cow 35 and normal-control 4 were negative."

Page 314, line 7, under heading "Discussion", "(*1, 19, 23*)" should be "(*2, 19, 23*)."

Page 314, twelfth line from bottom, "(*6, 7, 29*)" should be "(*7, 29*)."

Page 411, table 8, under Family 81.22, last column (difference) should read "2.0, 1.4, 2.3, .1, −.3, .4, −.2, − 2, −.5, .4, 0, −.4, .1, 0, −.7, and .1."

Page 419, line 5, "raw-cooked" should be "raw, cooked."

Page 419, footnote, reference number 4 to literature should be italic and enclosed in parenthesis.

Page 422, table 3, last line of heading, comma should be inserted after "*series 2.*"

Page 453, paragraph 3, line 5, "15 to 40" should be "15 to 25."

Page 453, fifth line from bottom, "typical" should be "special."

Page 458, table 3, second column from right under "First year", Desiccated meat meal, "5." should be "5.3."

Page 504, line 5, under "Backcross Data", "white" should be "yellow."

Page 516, figure 1, cells labeled "A" are described in legend "C" and vice versa.

Page 643, legend under plate 6 should appear under plate 7 and legend under plate 7 should appear under plate 6

Page 666, table 6, omit "Infected" in the two box heads.

Page 668, seventeenth line from bottom, "table 6" should read "table 7."

Page 675, table 1, last column, third number should be "15."

Page 752, figure 1. This illustration is upside down.

Page 833, the formula at the bottom of the page should be "Concentration (grams per 100 cc extract) $=35.51-\sqrt{1261-9.615\,(OR_a-OR_b)}$."

Page 841, lower right-hand corner, key no. should be "K-279."

Page 845, table 1, column 5, last line under Phenol, "^{3} 1,420" should read "$>^3$ 1,420."

Page 862, table 1, "cc units" should follow "Alkalinity of ash" and be deleted after "Vitamin A" and "Vitamin C."

Page 876, table 4, columns 4, 6, 8, and 10, "Ratio to total dry matter" should be "On basis of total dry matter." Columns 5 and 7, "Total dry matter insoluble in 70-percent alcohol" should be "On basis of dry matter insoluble in 70-percent alcohol." Columns 9 and 11, "Total dry matter soluble in 70-percent alcohol" should be "On basis of dry matter soluble in 70-percent alcohol."

Page 944, table 4, first line of heading, "loteners" should be "lateness."

JOURNAL OF AGRICULTURAL RESEARCH

VOL. 53 WASHINGTON, D. C., July 1, 1936 No. 1

EFFECT OF CARBON DIOXIDE ON THE CARBOHYDRATES AND ACIDITY OF FRUITS AND VEGETABLES IN STORAGE [1]

By ERSTON V. MILLER, *assistant physiologist*, and OSCAR J. DOWD, formerly *junior physiologist, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, United States Department of Agriculture* [2]

INTRODUCTION

The effect of carbon dioxide storage on carbohydrate transformation in certain fruits and vegetables was discussed in an earlier paper (*18*).[3] The present paper supplements the previous one by presenting data on three additional points: (1) Carbohydrate transformation in vegetables following removal from carbon dioxide storage, (2) carbon dioxide storage of several products not previously included, and (3) the effect of carbon dioxide storage on the acidity of certain fruits and vegetables.

The first point was suggested by inquiries regarding the earlier work. The question was raised as to how long treated peas and corn would maintain a high sugar content after removal from carbon dioxide storage and to what extent this advantage could be passed along to the ultimate consumer. The acidity studies were suggested by flavor tests conducted on fruits following removal from carbon dioxide storage. Several individuals found that there was a loss in sourness or astringency in the treated fruits.

REVIEW OF LITERATURE

The rapid deterioration in quality of freshly harvested fruits and vegetables has led to a study of the chemical changes that take place in these products and to an attempt to store them under conditions best adapted to maintenance of quality. Carolus (*7*) was able to retard the loss of total sugar in shelled green (Henderson Bush) lima beans by storing them at 0° to 2° C. Where the beans were held in cellophane bags, much of the total sugar was retained but the flavor was usually impaired. The sugar loss was retarded to some extent by holding the beans unshelled. Leaving them on the vine was more effective than the cold-storage treatment.

Several writers have shown that the quality of carrots deteriorates in storage. Hasselbring (*14*) found that the principal changes were a conversion of sucrose to reducing sugars and a transformation of polysaccharides to simple sugars. These processes occurred more rapidly at the higher temperatures, and under constant conditions tended to reach equilibrium after about 10 weeks. After this time there was a slight increase in sucrose. Platenius (*21*) reported similarly that sucrose in stored carrots was converted to reducing sugars at first but that the process was later reversed.

[1] Received for publication Dec. 2, 1935; issued August 1936.

[2] The writers are indebted to Charles Brooks, of the Division of Fruit and Vegetable Crops and Diseases, for helpful suggestions in planning these experiments, and to L. P. McColloch, of the same Division, for assistance in preparing the materials for analyses.

[3] Reference is made by number (italic) to Literature Cited, p. 16.

The possible effect of carbon dioxide on acidity was suggested as early as 1920, when Sando (*22*) noted that tomatoes, picked green and ripened without ventilation, showed high titratable acidity. He suggested that incomplete oxidation of carbohydrates to carbon dioxide was responsible for the accumulation of the acid.

In 1924 Magness and Diehl (*17*) treated Winesap and Delicious apples with various percentages of carbon dioxide for 10 days at 22° C. The total acidity disappeared more rapidly in the higher concentrations of carbon dioxide, the results being less pronounced for Delicious than for Winesap varieties.

Thornton (*23*) reported on carbon dioxide treatment of plants and various types of plant tissues, such as fruits, roots, stems, tubers, and bulbs, at 2°, 5°, 15°, and 25° C. Carbon dioxide in the presence of oxygen (30–60 percent CO_2 and 20 percent O_2) resulted in a decrease in hydrogen-ion concentration of the sap extracted from the tissue. Maximum differences were obtained at 25° during the period between 96 and 168 hours of storage. The reduction in hydrogen-ion concentration was found to be dependent upon the presence of oxygen in the gas mixture, and where this gas was lacking there was an increase in hydrogen-ion concentration.

Kidd and West (*16*), working with Lane's Prince Albert apples, report that increasing the concentration of carbon dioxide accelerated the loss of acid, but that reducing the concentrations of oxygen had little effect on acid loss. The concentrations of carbon dioxide employed were 5, 10, and 15 percent.

Hartman and Bullis (*12*) report no striking chemical or physical changes in stored cherries except losses of weight and volume, and state that there may be even a slight increase in the acidity of the juice if the cherries are held long enough. Nightingale, Addoms, and Blake (*19*) report an increase in acidity of Elberta and Shipper Cling peaches with development until the fruit reaches the soft ripe stage, when the percentage of acid suddenly decreases. Allen (*1*) found that, generally speaking, there is little significant change in acid content of peaches with maturity. A slight decrease continues after harvest, but this does not reach its minimum until the fruit has passed its prime eating condition.

The effect of low temperatures on flavor and subsequent ripening of mature green tomatoes has been discussed by a number of investigators. Diehl (*8*) stated that the exposure of mature green Globe tomatoes to a temperature just above the freezing point was not harmful if the exposure did not extend beyond 5 days. He also found that if mature green Acme, Stone, and Globe tomatoes were held at 32° F. for 8 days and then removed to a warm room they broke down and decayed without losing the green color.

Wardlaw and McGuire (*24*) were able to hold mature green tropically grown tomatoes at 47.5° F. for 20 days and then ripen them at 70°. Wright et al. (*25*) stored mature green tomatoes at 32°, 36°, 40°, and 50° for various periods and then transferred them to 60° to ripen. They found that the fruit ripened but failed to color properly if the preliminary storage consisted of 8 to 11 days at 32° and 36°, 11 days or more at 40°, or more than 14 days at 50°. Barker (*4*) found that the storage of tomatoes below 50° resulted in physiological injury. Haber (*11*) reported similar results. Sando (*22*)

reported that tomatoes picked green and ripened without ventilation were inferior in flavor and that coloring was retarded. Kidd and West (*15*) reported that carbon dioxide of higher concentration than 5 percent at 12° C. had an injurious effect on tomatoes.

MATERIALS AND METHODS

The peas, beans, corn, carrots, and plums used in these experiments were grown at Arlington Experiment Farm, Rosslyn, Va., near Washington, D. C. The peaches were obtained at Leesburg, Va., and transported to Washington by motor truck. Tomatoes and cherries were purchased from the local wholesale market. All materials were brought to the laboratory early in the morning and placed under experimental conditions as rapidly as possible. As a rule the experiment was started within an hour after the arrival of the fruit at the laboratory.

The experimental material was placed in 5-gallon glass pickle jars and held at 0°, 5°, 10°, 15°, and 20° C. Constant temperatures and constant gas percentages were maintained as previously described (*5*). In the latter half of these experiments a slight departure was made from the former method of mixing carbon dioxide with air. The air and the carbon dioxide were supplied directly to the mixing chamber by cylinders of compressed gases, the desired percentage and volume being obtained by means of a reducing valve and flow meters. An additional cylinder of compressed air was connected to the control lots and adjusted to secure a constant renewal of the storage atmosphere comparable to that of the treated lots.

If solid carbon dioxide is used as a supplementary refrigerant for fruits and vegetables the gaseous carbon dioxide may run as high as 40 to 50 percent during the first 24 hours, and it was the object of this investigation to study the biochemical effects of such a percentage of carbon dioxide on the product.

Large quantities of the fruits and vegetables were employed in order that they might be carefully culled and representative specimens selected for each lot. At the time of sampling the following quantities were removed from each lot: 500 g of peas, 500 g of lima beans, 500 g of cherries, 10 ears of sweet corn, 5 to 10 tomatoes, 10 peaches, 7 carrots, and 25 plums. These samples were thoroughly mixed, and 20 g and 25 g duplicates were preserved for carbohydrate determinations and 50 to 100 g for determinations of acidity.

The methods employed for sampling were essentially those prescribed for plants by the Association of Official Agricultural Chemists (*3*). Peas, beans, and corn kernels were dropped into sufficient hot 95-percent alcohol to make the final concentration of alcohol 80 percent. Peaches, tomatoes, plums, cherries, and carrots were minced in a food chopper before preserving in alcohol. The samples were boiled for 5 minutes and were then stored for subsequent analyses.

Official methods were employed for carbohydrate analyses. The fruits were extracted in a Soxhlet extraction apparatus with 80-percent alcohol until sugar-free. Peas, beans, corn, and carrots were dried after the preliminary extraction and ground to pass through a 40-mesh sieve before being transferred to the Soxhlet extractors.

Reducing sugars were determined by the Munson and Walker gravimetric method. Sucrose was determined as invert sugar after inversion with a commercial preparation of invertase that had been standardized with a Bureau of Standards sample of sucrose.

Moisture was determined by weighing 3 to 5 g of the mash, covering with absolute alcohol, and drying to constant weight in a vacuum oven at 65° C. and 15 inches of mercury.

Total acidity of the fruit juice was determined by straining the minced pulp through one thickness of cheesecloth and titrating aliquots with N/10 sodium hydroxide. Bromthymol blue was used as an indicator.

Carotene was determined by the method described by Palmer (*20, pp. 251–254*), with a Klett colorimeter for comparisons.

Hydrogen-ion determinations were made by means of a portable potentiometer, a quinhydrone electrode, and a calomel half-cell. The apparatus was standardized by means of a standard solution of acid potassium phthalate. Readings were also compared with those obtained with a bubbling hydrogen electrode, and it was found that the results recorded at the duration of 1 minute checked within 0.05 of a pH unit with the readings after equilibrium on the hydrogen electrode.

Total acidity and pH values on the total fruit pulp were obtained as follows: A 100-g sample of the minced material was covered with boiled distilled water and boiled for 1 hour. The mixture was made up to 500 ml when cooled to 20° C. This was filtered, and acidity and pH determinations were made on aliquots of the filtrate.

CARBON DIOXIDE AS A SUPPLEMENT TO REFRIGERATION

RATE OF COOLING OF LIMA BEANS AND SWEET CORN IN STORAGE

The importance of refrigerating green peas, lima beans, and sweet corn as soon as possible after harvest has frequently been stressed. One point, however, is rarely mentioned. There may be considerable lag in lowering the temperature of the vegetables to that of the storage room, both because of their high temperature at the time of harvest and because of the heat of respiration following harvest. Figures 1 and 2 illustrate this point. Henderson Bush lima beans (fig. 1) and Golden Giant sweet corn (fig. 2) were harvested in the forenoon, packed in bushel hampers, and placed in the 0° C. room at Arlington Farm at 1 p. m. Resistance thermometers were placed at various places in the pack, and the temperatures were read, without entering the room, by means of cables and a resistance box. As will be seen from the charts, the initial temperature of the vegetables was about 27°. During the first 2 hours the interior of the sweet-corn pack rose to 31.9°. After that the temperature began to drop. The temperature of the lima beans began to drop as soon as the beans were placed in storage. It is significant that it was nearly 20 hours before the temperature of the beans attained that of the storage room and 44 hours before the corn in the outer layers of the hamper attained this temperature. The center of the corn pack never attained the temperature of the storage room during the period of the observations.

IMPORTANCE OF EARLY TREATMENT WITH CARBON DIOXIDE

It is obvious, then, that since fresh vegetables under usual storage conditions are cooled down rather slowly, even when rushed into refrigeration as quickly as possible, the sugar content and freshness,

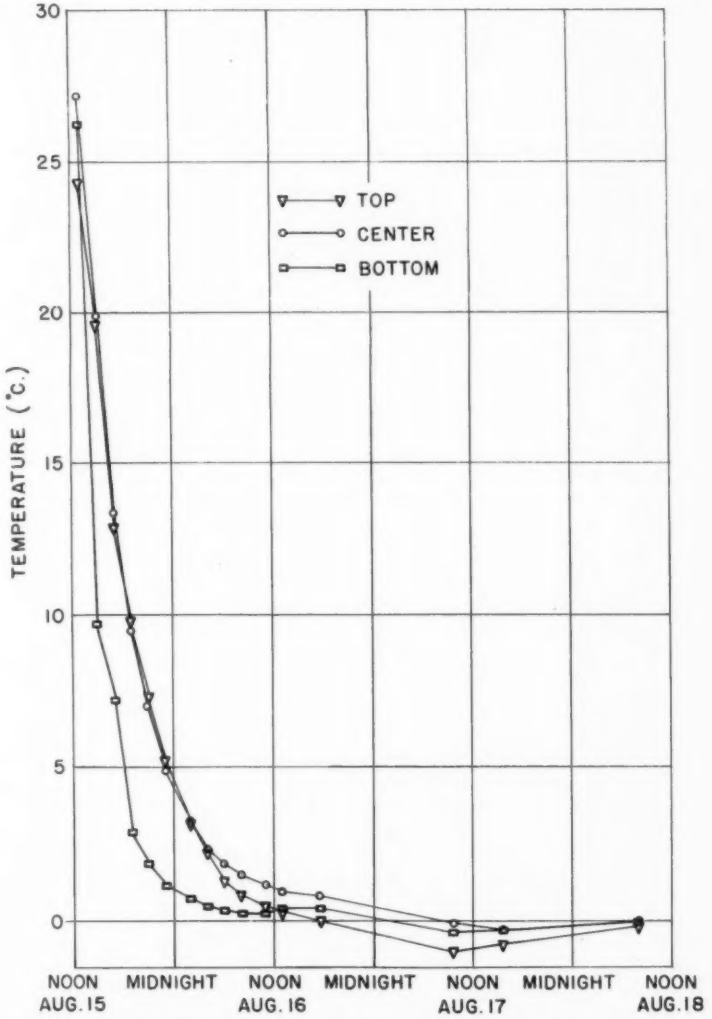

FIGURE 1.—Rate of cooling of Henderson Bush lima beans stored in bushel hampers at 0° C.

which are so desirable in certain vegetables, must be seriously diminished during the cooling process. For this reason it would seem advantageous to retard the respiration and sugar loss by producing an

atmosphere of carbon dioxide for the first 24 or 48 hours or until the pack is cooled to the desired temperature.

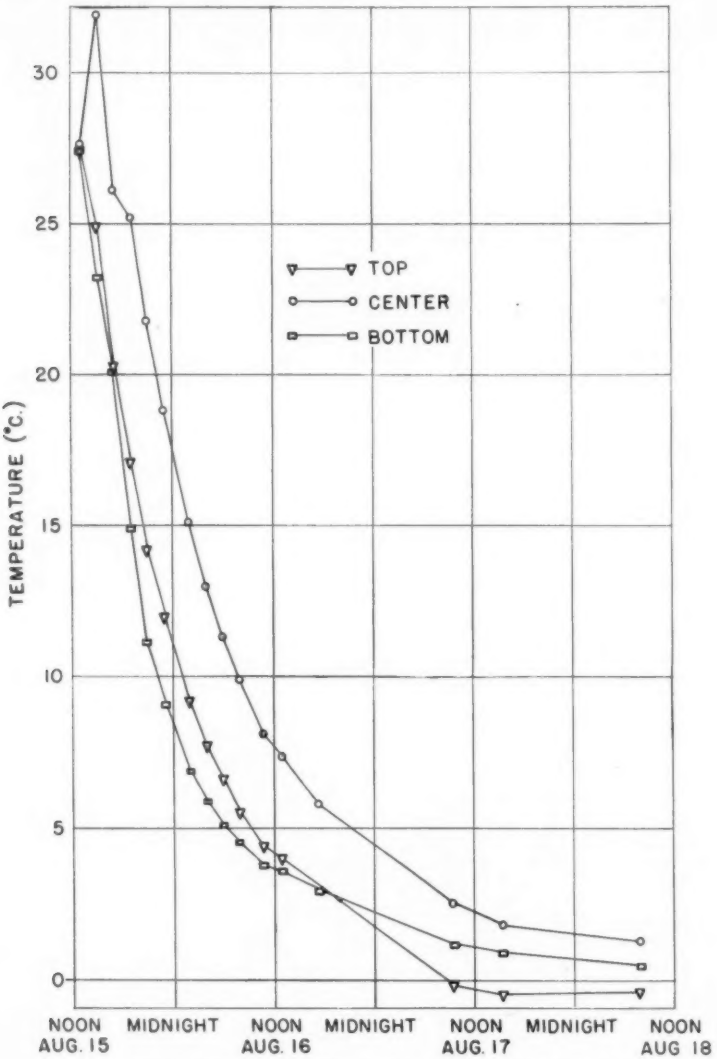

FIGURE 2.—Rate of cooling of Golden Giant sweet corn stored in bushel hampers at 0° C.

EFFECT OF CARBON DIOXIDE ON SWEET CORN CANNED IMMEDIATELY AFTER TREATMENT

As already mentioned, the sugar loss in sweet corn was retarded by carbon dioxide in storage experiments in the laboratory. In the summer of 1933 similar experiments were conducted at a canning

factory at Aberdeen, Md. Nine bushels of Country Gentleman (Shoe Peg) sweet corn selected at random from the supply reaching the factory early in the morning were mixed thoroughly and sorted into three equal lots. All three lots were stored immediately for 24 hours, as follows: Lot 1 was placed in a pony refrigerator [4] containing 10 pounds of solid carbon dioxide and 75 pounds of ice; lot 2 was placed in a pony refrigerator containing 100 pounds of ice; lot 3 was stacked on the floor of the canning shed. Gas determinations indicated that the carbon dioxide content of the atmosphere in the refrigerator containing lot 1 rose to 50 percent within 4 hours after the refrigerator was closed, and dropped to 37.6 percent when the refrigerator was opened after 24 hours' storage. Respiration in lot 2 raised the carbon dioxide content of the refrigerator atmosphere to 8.5 percent, 10 hours after the refrigerator was closed.

The outside air temperature at the beginning of the experiment was 23.3° C. The temperature of lot 1 had dropped to 10° by midnight, and that of lot 2 to 7.7°, while the center of the pile of lot 3 rose to 30°. At the time of opening, after 24 hours, the temperature of lot 1 was 6.9°, that of lot 2 was 6.1°, and that of the center of the pile of lot 3 was 27.7°.

The lots were removed from the refrigerators and from the stack on the floor, husked, cut from the ear, and canned immediately. Each lot was processed separately and the machinery was cleaned after each lot. In addition, a fourth lot of canned corn (lot 0) was reserved for flavor and sugar tests. This consisted of corn that had been canned immediately after arrival at the factory and was selected from straight factory run.

The canning process consisted in mixing a hot saline solution with the kernels, sealing, and processing for 40 minutes at 115.5° C. with 12 pounds' steam pressure.

All lots were taken to Washington, D. C., where they were tested for flavor and sugar content.

In order to subject the treated lots to as severe a test as possible, they were compared in flavor with that of the lot canned immediately (lot 0). Some of the persons testing the corn preferred the carbon dioxide treated lot, while others were able to detect something resembling an overcooked flavor in this lot. Rating all lots in the order of preference of the taster, and assigning a value of 100 to the lot that had been canned immediately, the others received the following values: Lot 1 (carbon dioxide), 72.8 ; lot 2 (ice), 70; lot 3 (stacked on floor), 47.1.

Fifty-gram samples were removed from cans of each lot, extracted with 80-percent alcohol, and analyzed for sugar according to official methods (*3*). Inasmuch as water had been added to the cans during the canning process, care was exerted to keep the contents well stirred while the sample was being taken. The variation in sucrose content of samples from duplicate cans in the same lot was never greater than 0.09 percent and averaged 0.05 percent. Comparing the total sugar in lots 1, 2, and 3 with that in lot 0 considered as 100, the percentages of sugar retained are as follows: Lot 1, 91.2; lot 2, 71.0; lot 3, 43.8.

[4] A pony refrigerator is an insulated box employed for shipping perishable fruits, such as strawberries, from southern production areas to northern markets before carlot shipments are moving. It contains a shallow ice pan beneath which is a compartment having a capacity of 64 or 80 quarts.

Two weeks later the experiment was repeated. The second experiment was similar to the first in all details except that the corn was stored for 42 hours instead of 24 and most of it had attained the late milk or early dough stage of maturity as compared with the early milk stage of the corn used in the first experiment.

In the flavor tests in this experiment a slight preference for lot 2 over lot 1 was shown. The ratings were as follows: Lot 1 (carbon dioxide), 72; lot 2 (ice), 74; lot 3 (stacked on floor), 53. Analyses for total sugar gave results similar to those in the previous experiment. Compared with lot 0 as 100, the percentages of sugar retained were as follows: Lot 1, 92.8; lot 2, 89; lot 3, 47.6.

IMMEDIATE EFFECT OF CARBON DIOXIDE STORAGE

CARBOHYDRATE TRANSFORMATION IN LIMA BEANS AND CARROTS

Henderson Bush lima beans stored in an atmosphere containing 42 percent carbon dioxide for 1 to 5 days reacted in a manner similar to that previously reported for peas (*18*). For the purpose of retaining the sucrose, a storage temperature of 5° C. was nearly as effective as 0°, and an atmosphere containing 42 percent of carbon dioxide made little difference at 5°. At 15° and 25°, however, the sugar loss was more rapid and the carbon dioxide was effective in retaining much of the sugar content (fig. 3). The lot stored at 25° with 42-percent carbon dioxide retained 69 percent of the original sugar on the first day and 51 percent on the second. The control lot at this temperature retained 43 and 31 percent on the first and second days, respectively. At 15° the carbon dioxide treated lot retained 86 percent of its original sugar on the first day and 79 percent on the second day. The control lot retained 68 percent of its sugar on the first day and 58 percent on the second. These differences in sugar content continued over the third day of treatment, but the carbon dioxide treated lots had lost their palatability on that day.

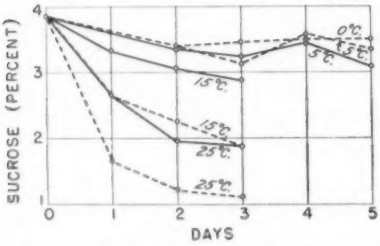

FIGURE 3.—Sucrose content of Henderson Bush lima beans stored in an atmosphere containing 42 percent carbon dioxide for 1 to 5 days at various temperatures. Solid lines, treated; broken lines, control.

Figure 4 shows the results of the carbon dioxide studies on carrots. The carrots received 39-percent carbon dioxide for periods of 3 days. The values represent averages of two identical experiments performed at different times with similar lots of carrots. The sucrose content decreased gradually at 15° and 25° C., but the rate of loss was retarded in the lots treated with carbon dioxide. The greatest contrast was observed on the second day. At this time the lot treated with carbon dioxide at 25° had 1.5 times as much sucrose as the control, and the similar lot treated at 15° had 1.4 times as much.

The reducing sugars tended first to increase and then to decrease in both the treated and untreated lots. In all cases the percentage of reducing sugar was higher in the controls than in the lots treated

with carbon dioxide. At 5° C. there was little contrast between treated and control lots in either sucrose or reducing sugars.

Additional lots were held in carbon dioxide beyond the 3-day period to test the effect on flavor. The limit of tolerance appeared to be 9 days at 5° C., 6 days at 15°, and 4 days at 25°.

EFFECT ON ACIDITY OF FRUITS

At the time of the original studies of the effect of carbon dioxide upon carbohydrates in fruits, acidity studies were made on cherries and peaches stored for 24 and for 48 hours at 0°, 5°, 10°, 15°, and 20° C. Early Richmond cherries received 50-percent carbon dioxide for 24 hours. Elberta peaches were exposed to 40-percent carbon dioxide for 24 and 48 hours. The results appear in tables 1 and 2. The carbon dioxide exerted no consistent effect upon the acidity of the cherries or the peaches during the period of treatment.

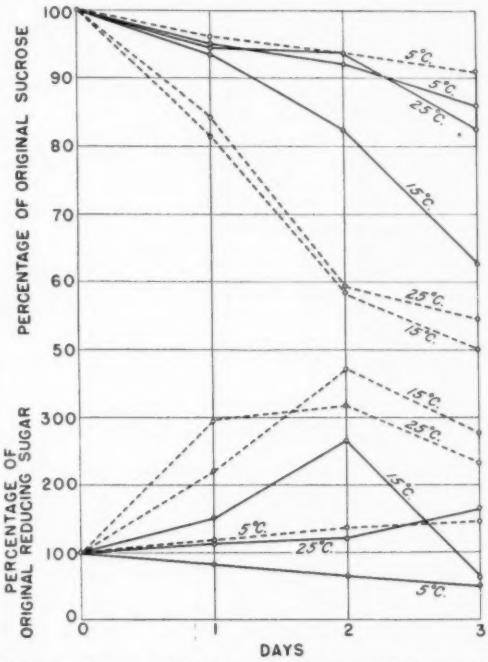

FIGURE 4.—Sucrose and reducing sugar in Danvers Half Long carrots stored in an atmosphere containing 39 percent carbon dioxide for 3 days at various temperatures. Solid lines, treated; broken lines, control.

TABLE 1.—*Total acidity and pH of Early Richmond cherries after 24 hours' treatment with 50-percent carbon dioxide at various temperatures*

Temperature (° C.)	Acid in fresh pulp as malic		pH of fresh pulp		Temperature (° C.)	Acid in fresh pulp as malic		pH of fresh pulp	
	Treated	Control	Treated	Control		Treated	Control	Treated	Control
	Percent	*Percent*				*Percent*	*Percent*		
[1]	[1] 1.37	[1] 1.37	[1] 3.47	[1] 3.47	10	1.50	1.45	3.41	3.40
0	1.45	1.42	3.45	3.50	15	1.47	1.33	3.44	3.45
5	1.44	1.47	3.43	3.40	20	1.33	1.47	3.46	3.44

[1] Original sample.

TABLE 2.—*Total acidity of Elberta peaches treated with 40-percent carbon dioxide for 1 and 2 days at various temperatures*

Temperature (° C.)	Days	Acid in pulp as malic		Temperature (° C.)	Days	Acid in pulp as malic	
		Treated	Control			Treated	Control
		Percent	*Percent*			*Percent*	*Percent*
		[1] 0.74	[1] 0.74	5	1	0.68	0.66
25	1	.59	.68	5	2	.66	.72
25	2	.71	.71	0	1		.70
15	1	.67	.66	0	2		.64
15	2	.66	.73				

[1] Original sample.

Marglobe and Earliana tomatoes were treated for 6 days with 50-percent carbon dioxide at temperatures of 0°, 10°, and 20° C. The results appear in figures 5 and 6. In general there was a gradual decrease in acid and hydrogen-ion concentration at the higher temperatures during storage, but the decrease was greater in the carbon dioxide treated lots. This reduction in acidity, due to the gas treatment, was more pronounced for the hydrogen-ion concentration than for the total acidity. It will be noted, for instance, that the hydrogen-ion concentration was lower in the treated lots than in the controls at both 20° and 10°, whereas the total acidity was lower in the treated lots at 20° only. There was practically no reduction in acidity in either the treated or untreated lots stored at 0°.

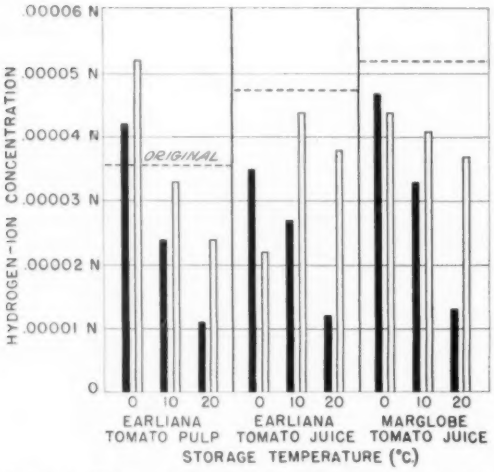

FIGURE 5.—Hydrogen-ion concentration of fruit pulp and fruit juice of Earliana tomatoes and fruit juice of Marglobe tomatoes treated with 50-percent carbon dioxide for 6 days at 0°, 10°, and 20° C. Shaded bars, carbon dioxide treated; plain bars, control.

Burbank plums treated with 50-percent carbon dioxide for 6 days at 0°, 10°, and 20° C. responded in a manner somewhat similar to that of tomatoes in regard to acidity changes. Figure 7 shows the percentage of titratable acidity in the juice after 3 and 6 days' treatment with 50-percent carbon dioxide. The results of two experiments have been averaged. The hydrogen-ion concentration showed a slight tendency to decrease with carbon dioxide treatment, but the results were not very consistent and were not considered significant. Differences in the total acidity are not great, but as a rule the carbon dioxide treatment seems to have reduced the acidity, especially at

temperatures of 10° and 20°. Because of too great a loss from decay, the 20° controls were not sampled on the sixth day. The lots treated with carbon dioxide at 20° were all sound. There was a slight rise in acidity between the third and the sixth day, but the treated lots were still lower in acid than the controls.

The Burbank plums were analyzed for benzoic acid content before and after treatment with carbon dioxide, but no significant differences were found.

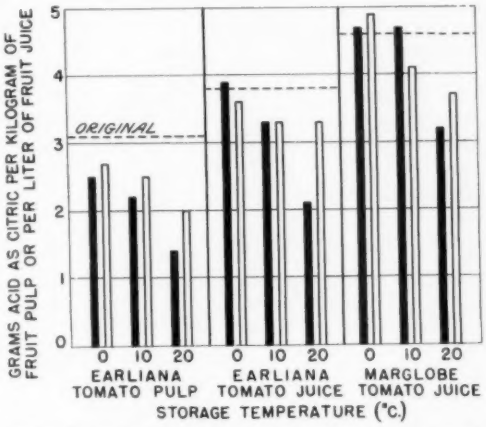

FIGURE 6.—Total acidity of fruit pulp and fruit juice of Earliana tomatoes and fruit juice of Marglobe tomatoes treated with 50-percent carbon dioxide for 6 days at 0°, 10°, and 20° C. Calculated on the basis of grams acid as citric per kilogram of fruit pulp or per liter of fruit juice. Shaded bars, carbon dioxide treated; plain bars, control.

GLYCOSIDES[5] OF PEACHES AND CARROTS

Peaches and carrots treated with carbon dioxide were analyzed for glycosides by Harvey's method (*13*) for phloridzin. Significant quantities of these substances were not obtained by the method employed.

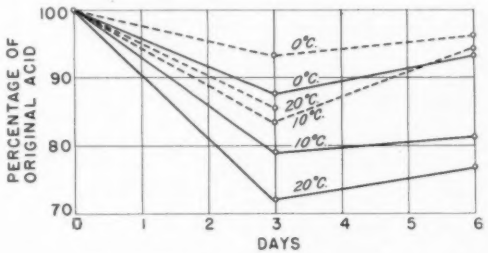

FIGURE 7.—Percentage of original titratable acidity in Burbank plums stored in an atmosphere containing 50 percent carbon dioxide for 6 days at 0°, 10°, and 20° C. Solid lines, carbon dioxide treated; broken lines, control.

CAROTENE IN CARROTS

In some of the earlier experiments the treated carrots were not as yellow as the untreated. Later, samples were analyzed for carotene content. Carrots were treated with 45-percent carbon dioxide for 11 days at 20° C., 14 days at 10°, and 15 days at 0°. At the end of these periods there was no difference between treated and untreated carrots in carotene content.

[5] The following definition is quoted from Armstrong and Armstrong (*2, p. 1*): "The term glycoside is applied to a large number of substances having the property in common of furnishing a sugar and one or more other products when hydrolised by acids * * *. The term glycoside is now used officially as a general name for the group, irrespective of the sugar present; glucoside is the specific name used for those glycosides the sugar constituent of which is glucose."

SUBSEQUENT EFFECT OF CARBON DIOXIDE STORAGE

CARBOHYDRATE TRANSFORMATION IN PEAS AND SWEET CORN

Figure 8 represents changes in sucrose content of Nott Excelsior peas stored in pods for 4 days at 0°, 5°, and 25° C. The lots treated at 5° and 25° received 42 percent carbon dioxide for 2 days and were then removed from the carbon dioxide chambers and held for the remaining time at the same temperatures in air. The controls were held in air during the entire period.

Both lots at 25° C. lost sucrose, but the control lot lost it much more rapidly than the treated lot. One day after removal from storage in 42-percent carbon dioxide, the treated lot retained 60.5 percent of its original sucrose as compared with 25 percent in the control. However, the 2-day treatment was too severe from the standpoint of flavor, and the treated lot eventually lost its palatability. At 5° the carbon dioxide treatment showed no advantage over the low temperature for retention of sugar.

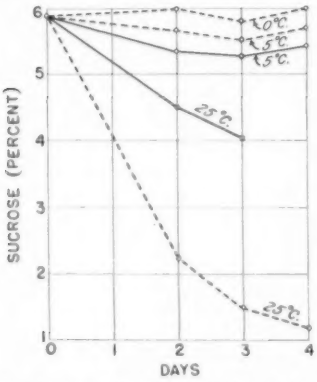

FIGURE 8.—Sucrose content of Nott Excelsior peas stored 2 days in an atmosphere containing 42 percent carbon dioxide and 2 days thereafter in air, at various temperatures. Solid lines, treated; broken lines, control.

Figure 9 shows the results of a similar treatment of Golden Giant sweet corn. Treatment with 42-percent carbon dioxide for 2 days so retarded the sugar loss at 5° C. that even up to the sixth day (fourth day after removal from carbon dioxide) the treated lot retained 74.6 percent of its original sugar as compared with 48.1 percent in the control.

At the end of the 2-day treatment the lot treated at 15° C. had 44 percent more total sugar than the control. On the fourth day the two curves practically coincided, but the control lot at this temperature had lost the bulk of its sugar during the first 2 days. The carbon dioxide treatment at 25° had no effect on retaining the sugar. The flavor was satisfactory in all cases in this experiment.

RIPENING OF TOMATOES

When the carbon dioxide studies of tomatoes were inaugurated it was noticed that some of the lots that received treatment appeared not to ripen properly. This was evidenced by the failure to obtain the full red color characteristic of ripeness. Additional studies of this phase were made, including total acidity determinations.

Marglobe tomatoes in the mature green stage were stored for 2 days in pony refrigerators under the following conditions: Lot 1, pan full of ice; lot 2, 25 pounds of solid carbon dioxide, and remaining space in the ice pans filled with ice; lot 3, 12 pounds of solid carbon dioxide, and remaining space filled with ice. A fourth lot was held in the ripening room (21° C.; 85–90 percent relative humidity) during the entire time. The carbon dioxide in lot 2 attained a maxi-

mum of 44 percent within the first few hours. The percentage of carbon dioxide dropped off gradually and registered 32.6 percent at the end of the second day, when the refrigerators were opened. During this same time the carbon dioxide in lot 3 ranged from 30 percent to 14.6 percent. After the refrigerators were opened, all lots were held in the ripening room for further observations. Ten representative tomatoes from each lot were reserved for notes on color changes. Every 3 or 4 days 10 tomatoes were removed from each lot and sampled for total acidity in the pulp.

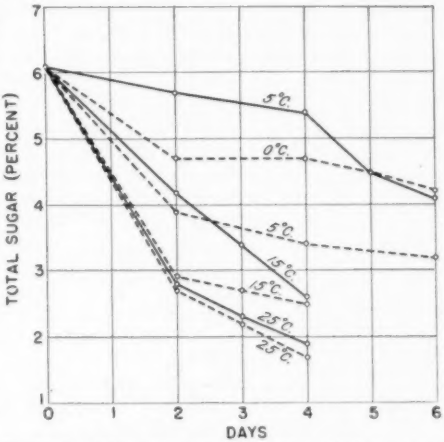

FIGURE 9.—Total sugar content of Golden Giant sweet corn stored 2 days in an atmosphere containing 42 percent carbon dioxide and 2 to 4 days thereafter in air, at various temperatures. Solid lines, carbon dioxide treated; broken lines, control.

Six days after the beginning of the experiment the lot stored in the ripening room showed 5 out of the 10 tomatoes red ripe and the rest turning or yellow red. None in the other lots had advanced beyond the turning stage. Ten days after the beginning of the experiment the tomatoes stored from the beginning in the ripening room were all red ripe or yellow red. Lot 1 and lot 3 had 6 and 7 red-ripe fruits, respectively, with the rest in the pink to turning stage. Lot 2, the tomatoes receiving the highest concentration of carbon dioxide, had 3 fruits in the yellow-red stage and the rest in the pink and turning stage. Three days later all save those in lot 2 were red ripe. Lot 2 never advanced beyond the yellow-red stage before

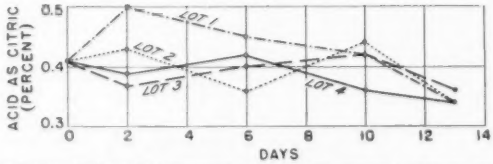

FIGURE 10.—Total acidity in Marglobe tomatoes stored under various conditions for 2 days and then stored in the ripening room (21° C.). Lot 1, stored 2 days in pony refrigerator containing ice; lot 2, stored 2 days in pony refrigerator containing ice and 25 pounds of solid carbon dioxide; lot 3, stored 2 days in pony refrigerator containing ice and 12 pounds of solid carbon dioxide; lot 4, stored in the ripening room (21° C.) for duration of experiment.

decay developed. Figure 10 shows the changes that took place in total acidity of the tomato pulp as a result of the different treatments. Following removal of the tomatoes from the refrigerators there was considerable fluctuation in the total acidity. At one time during the sampling period the acidity of each lot was higher than at the original sampling and at another time it was lower. At the

end of the 13-day storage period the acidity values were similar for all lots and had fallen below the original amount. Apparently the tomatoes were all physiologically ripe at the end of the experiment, even though pigment formation in lot 2 was altered if not inhibited. By the end of the experiment the flavor of all lots was considered fair.

DISCUSSION

The foregoing results indicate that the effect of storage in an atmosphere of carbon dioxide on peas and sweet corn is evident after removal from storage. However, the rate of sugar loss after removal is just as rapid in the lots treated with carbon dioxide as in the control lots, and in some instances it is more rapid, but the treated lots usually contain more sugar than the controls upon removal from the carbon dioxide. Thus the lots treated with carbon dioxide continue sweeter than the controls even though they are losing sugar rapidly after removal from the carbon dioxide treatment, and the advantage of the treatment is extended several days.

In this and in previous investigations it is shown that carbon dioxide has little effect on retaining sugar in sweet corn at 25° C. but a very pronounced effect at 5°, and that the reverse is true with peas and lima beans.

Green peas, sweet corn, and lima beans all lose their sugar rapidly at 25° C. Though similar in this respect, their response to carbon dioxide treatment indicates a physiological difference, and it would be interesting to determine whether this applies to other cereals and legumes.

The effect of carbon dioxide on carrots has been mainly to retard the conversion of sucrose into reducing sugar. Later in their storage life carrots begin converting reducing sugar into sucrose, but no attempt was made in these experiments to hold them until this stage was reached. In the present experiments the carrots treated with carbon dioxide were always sweeter to the taste and less coarse than the controls, when the carbon dioxide treatment was not continued too long. The flavor of the carrots was not affected by treatment for 4 days at 25° C., 6 days at 15°, or 9 days at 5°. Longer treatments at these temperatures usually produced a disagreeable flavor.

With the exception of peaches and cherries, the general effect of the carbon dioxide treatment on acids was to reduce the total acidity and hydrogen-ion concentration of the fruits and vegetables studied. This is in agreement with most of the work previously reported. The effect was usually most pronounced at the higher temperatures. The results were not always consistent for tomatoes at 10° and 0° C. It is probable that there is an effect of temperature that modifies any other effect. Under certain conditions storage of tomatoes at low temperatures prevents proper ripening of the fruit. In the experiments reported in this paper the period of storage may not have been long enough to interfere seriously with subsequent ripening. The normal physiological processes may have been sufficiently inhibited to produce the inconsistencies in acidity changes. Failure to color properly after removal from the refrigerators may have been due also to the high percentage of carbon dioxide.

A constant relation between total acidity and hydrogen-ion concentration was not always observed. In the studies on tomatoes the carbon dioxide treatment reduced the hydrogen-ion concentration much more consistently than the total acidity. The reverse was true of plums. Caldwell (*6*) states that there is no constant relation between total and active acidity values in fruits and vegetables, and only the broadest general parallel in their changes during ripening. The same result has been reported for vegetative parts by Gustafson (*10*). Diehl and Magness (*9*) found it difficult to obtain uniform samples for acidity determinations in plums. Allen (*1*) calls attention to the fact that the acidity in ripe plums becomes localized around the pit and in the flesh near the skin, which may influence the uniformity of the results.

Too much emphasis should not be placed upon the results on peaches and cherries as compared with those on tomatoes and plums, inasmuch as the former received but 24- or 48-hour treatments. But under these conditions there was apparently no reduction of acidity during treatment.

SUMMARY

The results of studies made to determine the effect of carbon dioxide upon fruits and vegetables are reported.

Green lima beans and sweet corn placed in storage at 0° C. immediately after harvest required 20 and 48 hours, respectively, to attain the temperature of the storage room.

Sweet corn held for 24 hours in 37- to 50-percent carbon dioxide in pony refrigerators retained about twice as much sugar as did that stacked on the floor during the same time, but after being canned it lost some of its original fresh flavor.

Loss of sucrose was retarded in lima beans and carrots by storage in approximately 40-percent carbon dioxide. The effect was much more pronounced at 15° and 25° than at 5° C.

Carbon dioxide treatment had no effect on acidity of Early Richmond cherries in 24 hours' treatment nor on Elberta peaches in 48 hours. Exposure of Earliana and Marglobe tomatoes to 50-percent carbon dioxide for 6 days resulted in a reduction of total acidity at 20° C. and a reduction in hydrogen-ion concentration at 10° and 20°. Treatment of Burbank plums with 50-percent carbon dioxide for 6 days resulted in reduction in total acidity at 0°, 10°, and 20°, but there were no consistent changes in hydrogen-ion concentration.

Peaches and carrots were analyzed for glycosides following carbon dioxide treatment, but no significant quantities of these substances were obtained. There were no significant differences in the benzoic acid content of treated and untreated Burbank plums.

Treatment of carrots with 45-percent carbon dioxide for 11 days at 20° C., 14 days at 10°, and 15 days at 0° did not reduce the carotene content.

Carbon dioxide had no permanent effect on carbohydrate transformation in peas and corn, except that for several days after exposure the treated lots still contained much more sugar than the controls.

Pony-refrigerator experiments with Marglobe tomatoes indicate that, although too high a concentration of carbon dioxide during storage may retard pigment formation during subsequent ripening,

the acidity changes are similar to those in the untreated lots and the reduction in acidity accompanying ripening is not inhibited by the carbon dioxide treatment.

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ABSORPTION, DISTRIBUTION, AND SEASONAL MOVEMENT OF POTASSIUM IN YOUNG APPLE TREES AND THE EFFECT OF POTASSIUM FERTILIZER ON POTASSIUM AND NITROGEN CONTENT AND GROWTH OF TREES[1]

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INTRODUCTION

In certain orchard investigations (*8*)[3] begun in 1928 at the University of Maryland, which involved a study of annual surface applications of potassium fertilizers to bearing apple and peach trees, no response in growth or fruiting was apparent even after several seasons. The question then arose as to whether there was an actual increased intake of potassium by fruit trees so treated under field conditions. The logical approach to the problem lay in comparing the chemical composition of trees treated with potassium fertilizers with those not so treated. To facilitate the study young trees were used. However, the same method of surface application of potassium fertilizer followed under ordinary field conditions was employed in the nursery row. Thus the results should be comparable to those of field studies with older trees. By studying the seasonal chemical changes in various fractions of these young trees, it was planned (1) to determine the period and rate of absorption of potassium; (2) to ascertain the relative concentration of potassium in the different parts of the tree, as well as the actual amounts present; (3) to obtain an indication as to any seasonal movements of potassium from one tree part to another; (4) to determine the effect of heavy applications of potassium fertilizer on the relationships stated above; (5) to study the effect of heavy applications of potassium fertilizer on the total nitrogen content of the tree parts; (6) to investigate the association of nitrogen with potassium in the tree parts throughout the season; and (7) to determine any growth responses caused by potassium fertilizer.

LITERATURE REVIEW

Working at New Hampshire in 1916 and using 7-year-old Golden Ball apple trees, Butler, Smith, and Curry (*5*) found that there was little movement of potassium until about the period of bloom, when there was a translocation from the older branches to the younger twigs. The young roots decreased in percentage of potassium from the dormant period until the time of bloom (May 18). From then until active growth ceased (July 12) the concentration increased

[1] Received for publication Aug. 13, 1935; issued August 1936. Part 2 of a thesis submitted to the faculty of the graduate school of the University of Maryland in partial fulfillment of the requirements for the degree of doctor of philosophy, 1934.

[2] Acknowledgment is made to the N. V. Potash Export MY, Inc. for providing certain funds necessary for this investigation; to Dr. A. L. Schrader, for reviewing the manuscript; to Dr. J. H. Beaumont for many kindly criticisms and suggestions; and to Dr. F. B. Lincoln for special assistance in planning the investigation.

[3] Reference is made by number (italic) to Literature Cited, p. 41.

markedly, and then fell off again. The leaves were not included in this study.

Richter (*13*), working in Germany about 1909, studied the potassium content of apple leaves throughout the growing season, and found that on a percentage basis the potassium content decreased as the season advanced. On an absolute basis, a maximum was reached at the stage of maximum leaf size. Roberts (*14*) in 1895, Van Slyke et al. (*19*) in 1905, and Thompson (*18*) in 1916 all attempted to interpret the fertilizer needs of fruit trees by chemical analysis of the entire tree and a calculation of the total amount of any essential element removed over a definite period. Their work yielded no additional information as to seasonal movements of potassium, but it did give an indication as to where potassium was located in the apple tree. Since Roberts worked with a nonbearing tree, even though it was mature, some of his data recalculated to show the relative proportions of the potassium located in the different parts of the tree, are presented. The analysis showed that 15.0 percent of the tree's potassium was located in the leaves, 7.1 percent in the twigs of the current season, 9.0 percent in the 1-year twigs, 58.3 percent in the trunk and limbs, and 10.5 percent in the roots.

Burke and Morris (*4*) in Montana studied the movement of potassium from the tree into the new growth and leaves of 20-year-old apple trees. One tree was sampled during the dormant period (Apr. 9) and another when the leaves had attained full size (June 11). By comparing the amount of potassium in the dormant tree with that in the active tree they could calculate the extent of movement up from the soil and roots, as well as the movement from the old wood into the new growth and leaves. They found that potassium moved upward from the roots, trunk, and large limbs. There was sufficient movement from the soil and roots, however, to supply the needs of the leaves and new twig growth. Unfortunately, they did not dig the entire root system and therefore could not differentiate between potassium coming from the soil and that coming from the roots.

MATERIALS AND METHODS

DESCRIPTION OF PLOTS

From a group of 400 2-year-old Stayman Winesap apple trees growing in the nursery row, on a Sassafras sandy loam at College Park, Md., 190 of the more uniform [4] were selected for the experiments. The block of trees consisted of 4 rows about 4 feet apart, the trees being approximately 18 inches apart in the row. The trees were allowed to remain in their original places.

The block was divided into four plots, each of which received an application of one-fourth pound of ammonium sulphate and one-half pound of superphosphate per tree. Two of the plots received an application of 1 pound per tree of sulphate of potash. This was considered to be a heavy application of potassium.[5] The materials were applied April 1, 1933.

[4] The coefficient of variability of the diameters of the 190 trees was 1.72 percent.

[5] Replaceable potassium determinations of the surface 18 inches of soil were made before the fertilizers were applied and at intervals throughout the growing season. The results indicated that the normally treated plots remained at about 34 p. p. m., while the potassium-treated plots increased to about 540 p. p. m. after treatment and remained at approximately that figure throughout the season.

The trees had been planted on a gentle slope with the rows set in the direction of the slope. The plot boundaries were perpendicular to the rows, so that each plot contained a section of all rows. Since not all trees in the rows were used, the number of trees in the plots was unequal. However, the number present in each plot was sufficient to be representative of the treatment. The plots were numbered from 1 to 4, going up the hill, 1 and 3 receiving the potassium.

GROWTH RECORDS

Since it was desired to study the effect of the heavy application of potassium upon the growth of the trees, records were taken at weekly intervals during the greater part of the growing season, although occasionally a longer interval between measurements was necessary. The diameter at a marked point was taken on each measuring date, the accuracy of the measurement extending to 0.01 cm. The longitudinal growth of the terminal shoots of each tree was measured in centimeters.

SAMPLING METHODS

Samples for chemical analysis were secured at 12 intervals between April 8, 1933, and January 8, 1934, inclusive. Since the trees had not previously exhibited any potassium-deficiency symptoms, the treatment involving only nitrogen and phosphorus was considered to be normal, and since a study under normal conditions was desired, four trees were taken as a composite sample from these plots on each sampling date. From the plots receiving a heavy application of potassium only a single tree was sampled on each date, the assumption being that if any marked compositional differences between the potassium-treated and the untreated trees existed, they could be detected by the analysis of the one tree. Furthermore, it was believed that the variability among the normally treated trees probably would represent the variability which would be expected from the trees receiving the potassium fertilizer. The trees dug on any sampling date were always selected at random, except that no two adjacent trees were used for analysis, thus avoiding any possible injury to the root systems.

The entire trees were dug, care being taken to preserve as many roots as possible. However, during the latter part of the season, when the root systems were rather extensive, some of the finer roots were lost. The trees were dug between 7 and 8:30 a. m., following a sunny day. They were never dug when rain was falling, although in a few instances rather cloudy weather prevailed. The digging was done as rapidly as possible and the trees were carried to the laboratory for subsequent treatment.

After the trees had been washed and weighed, they were separated into 18 different fractions. (There were fewer fractions during the early part of the growing season or when no leaves were present.) The following fractions were secured: Leaves, 1933 upper bark, 1933 upper wood, 1933 lower bark, 1933 lower wood, 1932 upper bark, 1932 upper wood, 1932 lower bark, 1932 lower wood, 1931 upper bark, 1931 upper wood, 1931 lower bark, 1931 lower wood, large root bark, large root wood, small root bark, small root wood, and rootlets.

The fractions were designated in accordance with the year of their first longitudinal extension. The upper and lower classification

designated the upper and lower half of each section. The large roots were those 1 cm or more in diameter; the small roots ranged from 1 cm to 2 mm in diameter; the remainder of the root system was designated as rootlets.

Since considerable time was required to fraction five trees, the bark/wood ratio was obtained from the tree fractioned for moisture determinations. (See under Chemical Methods.) Then the weights of the fractions, including bark and wood for all trees, were recorded and the dry weights of the individual fractions later were calculated from the bark/wood ratio and moisture content of the one tree.

Smaller samples, taken from the composite sample of the four trees from the no-potassium plots, were peeled, dried, and ground for chemical analysis.

All leaves were picked off and thoroughly mixed. From this lot about 200 leaves were selected at random to constitute the sample for analysis.

CHEMICAL METHODS

Moisture.—On the first three sampling dates the moisture content of all fractions of trees from both treatments was obtained, but inspection of the results showed that the trees sampled first always had a slightly higher moisture content, regardless of treatment. Therefore, on subsequent sampling dates the one tree from the high-potassium plots was immediately cut up for moisture determinations, so as to avoid unnecessary water losses. All moisture calculations were based on this one tree. The samples were dried at 72° C for 48 hours in a forced-draft oven and loss in weight was called moisture.

Nitrogen.—Total nitrogen was determined by the official Kjeldahl-Gunning method (*2*).

Potassium.—Two-gram samples of the ground, oven-dried material were ashed in an electric muffle furnace at 550° C. The ash was taken up with hydrochloric acid, transferred with water to a 100-ml volumetric flask, made to volume, and a 25-ml aliquot was taken. Potassium was then determined by the sodium cobaltinitrite method of Schueler and Thomas (*16*). Since the method used had not been accepted by chemists in general, it seemed advisable to check it with the official platinic chloride method. Through the courtesy of Dr. Lilleland, of the University of California, samples of prune leaves on which determinations had been run by the official method were obtained for analysis. Table 1 gives a comparison of the two methods as evidenced by the potassium content of the prune leaves.

TABLE 1.—*A comparison of the total potassium (as percentage of dry matter) content of prune leaves as determined by the platinic chloride and sodium cobaltinitrite methods*

Sample no.	Platinic chloride method	Sodium cobaltinitrite method
	Percent	*Percent*
1	1.46	1.50
2	1.31	1.29
3	2.31	2.38
4	2.85	2.99

The figures are in rather close agreement, considering the fact that different glassware, solutions, and operators, as well as different chemical methods, were employed in obtaining the results.

EXPERIMENTAL DATA

ABSORPTION OF POTASSIUM BY THE TREES

Since some variability existed among the trees, it was necessary to smooth out the dry weights of all fractions by means of a three-figure moving average. Since a moving average results in a reduction in the number of figures, the first two sampling dates were averaged for the first figure and the dry weight for the last sampling date was left unchanged; that is, the first corrected dry-weight figure was an average of the first two, the second corrected figure was an average of the first three figures, the third an average of the second, third, and fourth figures, etc. The next to the last corrected figure was an average of the last three, and the last corrected figure was the same as the last actual dry-weight figure. After the corrected dry-weight figures were obtained they were multiplied by their corresponding potassium percentages, which gave the actual grams of potassium present in each fraction (table 2).

TABLE 2.—*Total amount of potassium per tree under the two fertilizer treatments, both including and excluding the leaves, 1933*

Date sampled	Potassium-treated [1] [2]		No potassium [2]		Physiological state of trees
	Including leaves	Excluding leaves	Including leaves	Excluding leaves	
	Grams	*Grams*	*Grams*	*Grams*	
Apr. 8	0.9348	0.9348	0.8669	0.8669	Buds beginning to swell; bark sticking.
Apr. 25	1.0781	.8448	.9945	.7612	Terminal buds bursting; some leaves ¼ inch long; bark still sticking on roots.
May 11	1.5019	1.0542	1.4908	.9726	Buds fully opened; in some cases very slight terminal growth.
May 29	2.6898	1.5998	2.0502	1.2462	New growth 18–22 cm long.
June 13	4.8985	2.4695	2.6993	1.5120	Terminal growth 25–40 cm; lower part of 1933 growth turning brown.
June 25	----------	----------	3.5938	1.8541	Terminal growth 38–48 cm; trees still growing rapidly.
July 23	6.0350	2.8332	3.5368	1.9818	Terminal growth 50–70 cm; trees healthy and vigorous.
Aug. 18	5.8268	3.2275	4.0307	2.3055	Bark sticking on few upper twigs; growth slowing down.
Sept. 10	6.5827	4.0454	4.2655	2.6027	Some leaves falling; bark sticking on all parts of tree except trunk and roots.
Oct. 4	6.0751	4.1834	3.1832	2.3303	Many older leaves fallen; bark sticking on all parts of tree.
Nov. 3	5.2896	4.4887	3.6208	3.1518	Only a few of younger leaves left; some still green.
Jan. 8	----------	4.3528	----------	3.2686	Tree entirely dormant.

[1] Treated with 1 pound potassium sulphate in April.
[2] All trees received a basic application of $(NH_4)_2SO_4$ and superphosphate.

It happened that the four trees of the sample taken from the normally treated plots on October 4 were rather small. Even though their low dry weights were increased by the moving average, the amount of potassium in the trees was still low, indicating that the concentration was lower than would be expected at this time of the year. The potassium values obtained on October 4 are not plotted on the graphs because the trees were considered as abnormal.

No potassium-treated tree was dug on June 25.

In connection with table 2, a note as to the approximated physiological status of the trees at each sampling date is appended. When comparisons are made between the results of this investigation and those of similar studies, the use of the tree condition, rather than the calendar dates, would seem advisable. However, in this report, for the sake of brevity, all references to time of sampling are made in terms of the actual date. Since the sampling dates were arbitrarily

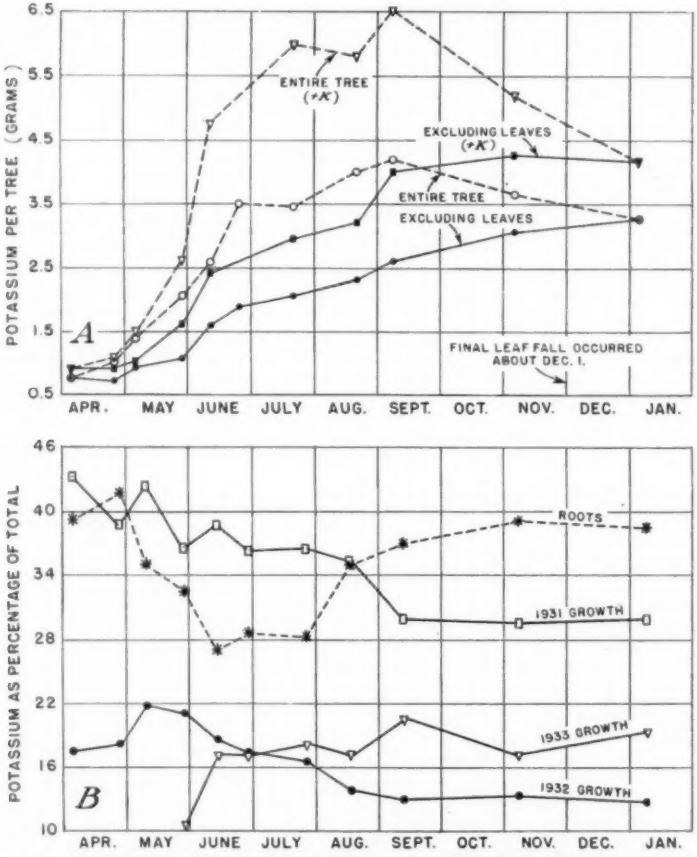

FIGURE 1.—Potassium content of: *A*, Trees, including and excluding leaves, throughout the season, under two different fertilizer treatments, with and without potassium. *B*, Different-aged portions of trees throughout the season. +K treated with 1 pound potassium sulphate in April; others untreated.

chosen without reference to any expected changes in potassium behavior, any difference between two dates may not represent the extent of the actual change which occurred.

Table 2 shows the total grams of potassium in the apple trees on the various sampling dates. Figure 1, *A*, presents the same material graphically. The data are given both for the entire tree and the tree excluding the leaves, so that the potassium content of the tree can

be studied without the introduction of the rather large amount of potassium which accumulates in the leaves, a large part of which is lost at leaf fall.

Considering both fertilizer treatments, it is evident that potassium was absorbed slowly between April 8 and 25. A portion of the potassium in the woody part of the tree moved into the leaves during this period, as is indicated by the fact that the potassium content of the tree, minus the leaf content, decreased.

From April 25 to May 11, the intake of potassium was sufficient to cause an increase in all parts of the tree, and the rate of absorption continued rather uniformly from May 11 to September 10. Leaf fall was beginning by the latter date, and from then on the potassium content of the tree as a whole decreased. However, excluding the leaves, the potassium in the tree continued to increase at a uniform rate until November 3. This increase may have resulted from autumnal migration from the leaves, but limited data which will be presented later indicate that the probability of absorption from the soil is greater. From November 3 until January 8 there was little change.

The absorption curve for the potassium-fertilized trees (+K) was similar to that for the normal treatment except that considerably more potassium was absorbed over the same period of time. Considering the large differences in potassium intake and the lack of differences in growth response of the two groups of trees, there can be no doubt that luxury consumption of potassium occurs with the apple.

SEASONAL MOVEMENT OF POTASSIUM IN DIFFERENT TISSUES AND PARTS OF TREE

In considering the seasonal movement of potassium in the different portions of the tree, each of the 18 fractions might be considered individually. However, to simplify the problem, only the different-aged portions of the tree are considered separately.[6] The fact cannot be disregarded that the growth laid down the previous season, and designated here as 1932 growth, has within it new tissues added in 1933. These tissues are probably of entirely different composition from those formed the previous year. Therefore, if tissues decrease in potassium with age and young tissues high in potassium are being deposited, the interpretation of the results is somewhat difficult. Some experimental evidence obtained on this point will be presented later. Meanwhile, the data for the potassium content of the tree fractions will be presented, disregarding the fact that the 1931 wood, for example, contains new wood added in 1932 and still younger tissues laid down in 1933.

The question arose as to whether the percentage of fresh-weight or the percentage of dry-weight figures should be used in interpreting the relative potassium concentrations in the different fractions. An inspection of the fresh- and dry-weight figures for the entire tree indicated that the dry weights presented a more stable basis on which to calculate the results. Dry matter accumulated constantly throughout the season. The fact that the fresh weights fluctuated considerably, even though the general trends were similar, indicated changes

[6] The complete data for the individual fractions are on file at the Maryland Agricultural Experiment Station.

in moisture content during the different periods rather than any appreciable change in dry matter.

Another problem which arose in connection with the study of the potassium status of the fractions was how to consider the leaves in connection with the other tissues. They are not a permanent part of the tree, are extremely vegetative, and contain a large proportion of the total potassium in the tree. Because of these conflicting features, the leaf data are presented separately.

Proportionate Distribution

Figure 1, *B*, presents graphically the potassium content of the different aged portions of the trees, expressed as percentage of the total. These data give an indication as to what portions of the tree contain the bulk of the potassium on the various sampling dates.

The roots at first carried 40 percent of the total potassium, but by June 13 they carried only 27 percent. In October they contained about the same proportion of the total that they had at the beginning of the growing season.

There was a general tendency for the 1931 and 1932 growths to lose in their proportionate amounts of potassium from May until September.

The 1933 growth tended to increase in its proportional amount of potassium. This might be expected since new tissues were being added throughout the season.

Averages for the entire season show that the bark of the tree under the normal treatment contained 47.5 percent of the total potassium, while the wood contained 52.5 percent, but under the potassium treatment 57.0 percent of the potassium was in the bark and only 42.1 percent in the wood. This would indicate that luxury consumption takes place more readily in the bark than in the wood. Other figures will bring this out more emphatically.

Absolute Distribution

Considering the same material expressed in grams per fraction, the actual movement of potassium from one part of the tree to another can be traced. These data, presented in figure 2, *A*, show that the different-aged parts of the tree continually gained in potassium. The roots did not increase in potassium during the period of early growth activity at a rate similar to that of the other fractions, but from July 23 on through the season their potassium content increased greatly.

The fact that the potassium content of the 1933 growth increased more rapidly than that of the 1932 growth might be attributed to the greater proportion of young tissue in the 1933 growth. Dowding (*9*) and many others have reported a high concentration of potassium in the younger tissues. This theory, however, does not hold true when the 1931 growth is considered, since its increase is more rapid then either of the younger tissues.

The total amount of potassium in the 1931 and 1932 growth followed closely the dry weight, as figure 2, *A*, and *B*, shows. The roots and new growth, however, increased in potassium during the latter part of the growing season more rapidly than they accumulated dry matter.

Concentration

The amount of potassium in each fraction was expressed as percentage of the dry matter. The results are shown in figure 3, *A*. The data indicate that the concentration of potassium in the new growth was very high at the start of the season as compared with that in other portions of the tree, but it decreased suddenly between May 29

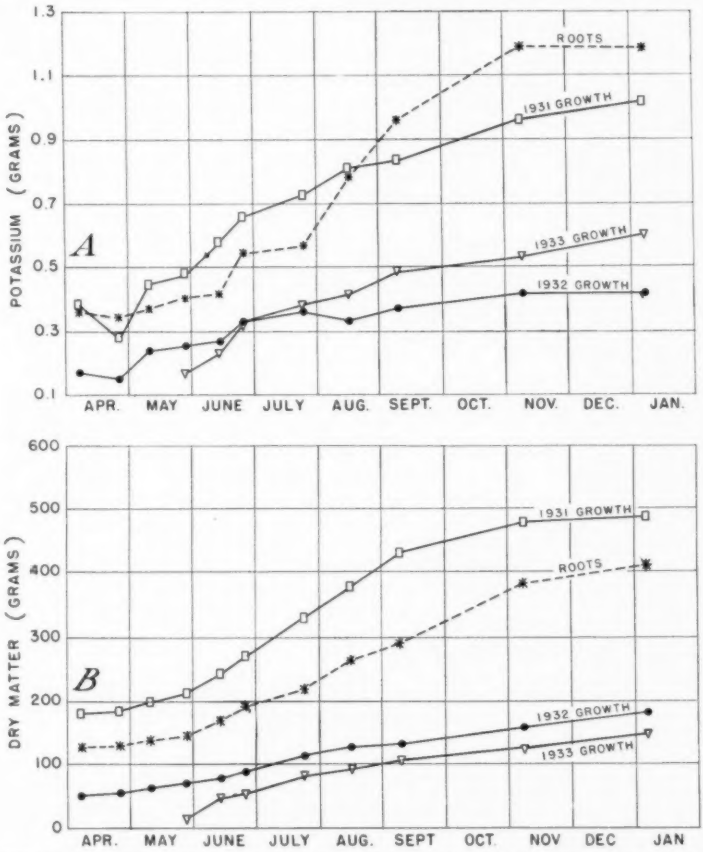

FIGURE 2.—Absolute amount of potassium (*A*) and of dry matter (*B*) in the different aged portions of the trees throughout the season.

and July 23. After that it decreased gradually throughout the growing season. The concentration of potassium in the 1932 growth, after May 29, showed a gradual decrease throughout the season, while that in the 1931 wood remained fairly constant, decreasing only slightly.

The potassium content of the roots remained rather constant until the latter part of July, when it increased. The increase continued until September 10, and the potassium remained high throughout the rest of the sampling period.

It is evident that the percentage composition is an expression of the relation between the absolute increase in potassium and the increase in dry matter. If dry matter and potassium increased proportionately, the percentage composition would remain the same. However, as the graphs bear out, in the roots the increase in potassium was greater than the increase in dry weight, while in the 1933 growth the reverse was true.

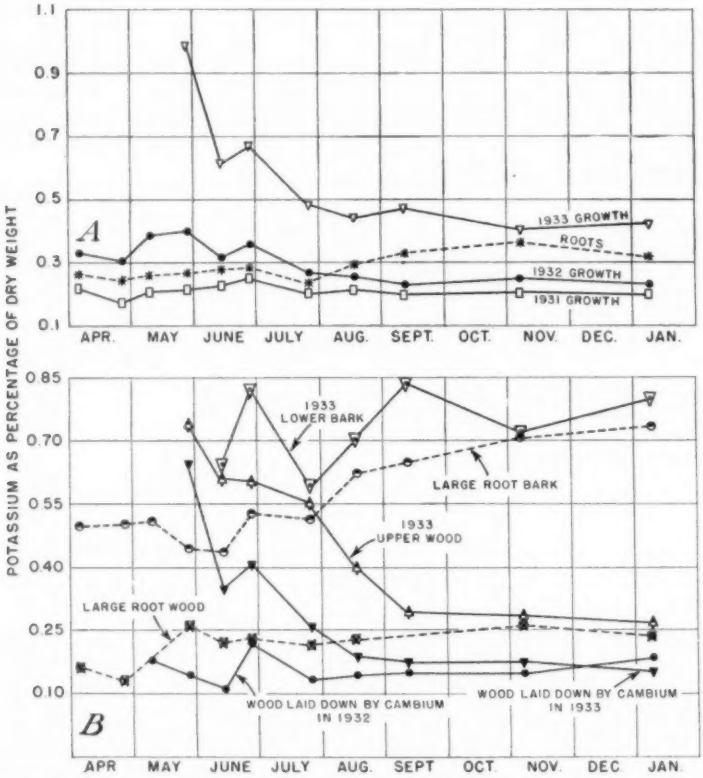

FIGURE 3.—Potassium content of: *A*, Different-aged portions of the trees; *B*, certain bark and wood fractions.

DISTRIBUTION BETWEEN BASAL AND APICAL PORTIONS OF GROWTH AND AMONG TISSUES

The question now arises, did both basal and apical portions of the different-aged parts of the tree act similarly throughout the season? Certain unpublished data indicate that, on a percentage basis, the bark and the wood, both upper and lower, of the 1932 and 1931 growth did act very similarly, but in the other parts differences existed.

The trends for potassium, as percentage of dry weight of the upper 1933 wood and lower 1933 bark, as well as those for the large root bark and root wood are presented in figure 3, *B*.

The large increase in the roots after July 23 was caused principally by increases in the bark rather than in the wood. The root bark and wood seemed to exhibit a somewhat reciprocal relationship during the early part of the growing season, thus accounting for the constant percentage composition of the roots as a whole. The causal factors are unknown. Figure 3, *B*, brings out the fact that the decrease in percentage composition of the 1933 growth was due largely to decrease in the wood fraction, since the bark showed no material decrease.

On an absolute basis the bark and wood fractions, both upper and lower, maintained trends similar to those of the entire season's growth (bark and wood, upper and lower combined), the only differential response being a relatively faster increase in potassium in the bark than in the wood. These statements apply to all sections of the tree regardless of age.

Certain unpublished data indicate that the old wood is relatively inactive with respect to potassium changes. Of course, if potassium

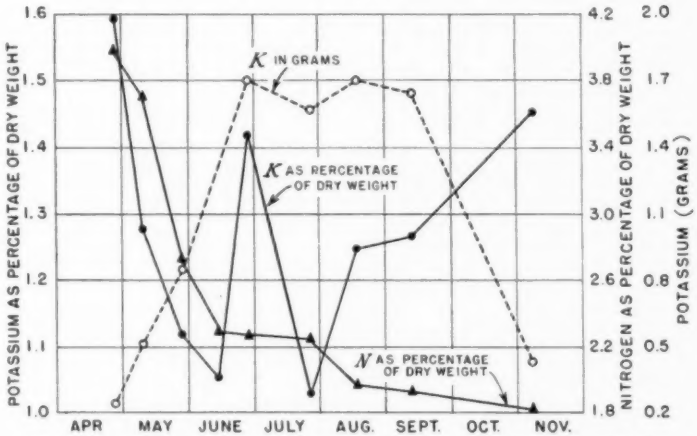

FIGURE 4.—Potassium content of the leaves throughout the season; also nitrogen content.

were merely passing through the conducting elements of the xylem, marked seasonal differences would not be expected. However, the new wood laid down by the cambium should be relatively high in potassium if the number of living cells in a tissue is correlated with its potassium content. In order to study this point, the new wood laid down on the lower 1932 section of the tree was separated on each sampling date and subsequently analyzed for potassium. The curves, as percentage of dry matter, are plotted in figure 3, *B*.

The curve for the new wood on lower 1932 growth (wood laid down by cambium in 1933) very closely resembles that for the upper 1933 wood. This appears to indicate that the decrease in concentration of potassium in new wood was similar whether it was initiated from the cambium or was a combination of an apical meristem and the cambium. The old wood laid down in 1932 did not show any marked change in potassium concentration throughout the season.

POTASSIUM CHANGES IN THE LEAVES

The leaves, at their maximum weight, contained 44 percent of the total potassium in the tree. The fact that the seat of photosynthetic activity is in the leaves may be associated with this high concentration, since potassium is considered to play a part in carbohydrate metabolism.

Figure 4 shows the potassium composition of the leaves on a percentage dry-weight and absolute-amount basis throughout the season, and the nitrogen composition as percentage of dry weight.

The curves indicate that on a percentage basis the potassium content of the leaves fell rapidly until June 13, rose sharply until June 25, then declined until July 23. From that time until leaf fall the concentration of potassium increased. The sudden rise between June 13 and June 25 cannot be explained. It is doubtful whether sampling error could account for it. The chemical determination was checked carefully.

On an absolute basis the potassium content increased until June 25, remained fairly constant until September 10, and then fell off as leaf fall began. The curve is such as one would expect since it conforms quite well with the total dry weight curve of the leaves.

The curve for nitrogen content in the leaves expressed as percentage of dry weight presents a picture quite different from the corresponding curve for potassium. The concentration of nitrogen decreased continually throughout the season, although rather slowly during the latter part of the growing period. The fact that the potassium concentration increased so strikingly during the autumn when the nitrogen concentration decreased, suggests the unlikelihood of an autumnal migration of potassium into the bark and wood. However, the fact that the older leaves fell before the younger ones tended to cause the potassium and nitrogen content of the leaves to be high at the close of the season and may have obscured some autumnal migration.

To substantiate the fact that young leaves contain more potassium than older ones, on three sampling dates the leaves growing on the upper end of the current season's growth were separated from those growing on the lower portion. The results are reported in table 3.

TABLE 3.—*Potassium content of old and young leaves at 3 sampling dates, 1933*

Date	Leaves	Potassium as percentage of dry matter	Potassium per 100 leaves
		Percent	*Grams*
July 23	Young	1.539	0.529
	Old	1.102	.337
Aug. 16	Young	1.632	.614
	Old	1.320	.459
Sept. 10	Young	1.639	.590
	Old	1.293	.383

RELATIVE POTASSIUM CONTENT OF THE VARIOUS FRACTIONS

Obviously the various fractions differed in their potassium content on the different sampling dates. However, certain relationships among the different parts held true generally throughout the season.

Figure 5 gives the average potassium content of the various fractions, samples from all dates being averaged.

On a percentage basis the leaves were the richest in potassium. As percentage of dry matter, there was a decreasing gradient of potassium in the bark from the apical end of the tree to the smallest roots. In the wood there was a similar decrease in potassium until the base of the tree was reached, but there the large roots showed

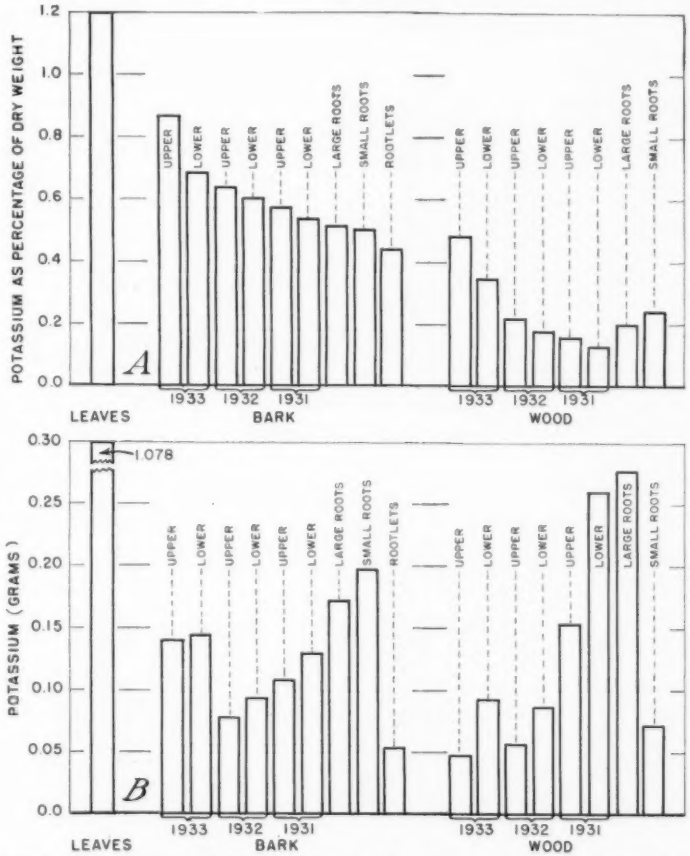

FIGURE 5.—Potassium content of the different fractions of the trees expressed as: *A*, Percentage of dry weight; *B*, as grams per fraction. Averages for entire season.

some increase in potassium concentration and the small roots even more.

A comparison of percentage results with the absolute amounts of potassium in the tissues (fig. 5, *B*) shows the leaves to be higher on both bases. With the exception of the 1933 bark, there was an increasing gradient of potassium on the absolute basis down the tree to the small root bark. Thus the absolute amounts were generally

negatively correlated with the percentage composition. The wood generally exhibited an increasing amount of potassium from terminal portion to root portion of the trees. The small roots were exceptional, and the 1933 lower wood was somewhat higher in potassium than the 1932 wood. However, in general, both with bark and wood, where the concentration of potassium was highest, the absolute amounts

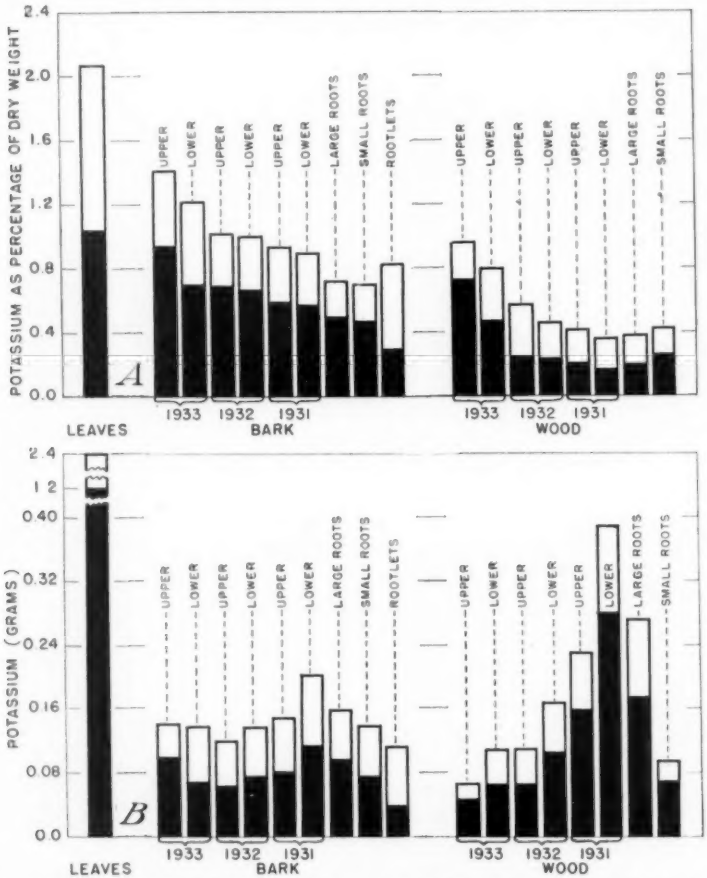

FIGURE 6.—Potassium content of the various fractions of trees on June 13, comparing: *A*, The potassium-treated trees with the normally treated trees; *B*, the treated trees with the normal ones. The black part of the bars denotes potassium content under normal treatment; the white part shows accumulation due to fertilizer application.

were lowest. The most logical explanation of this would be that potassium is highly concentrated in young, active tissues, while the bulk of the tree, being made up of woody structural material, has a low concentration of potassium. This point has not received proper consideration in the literature, many investigators reporting that most of the potassium was located in the bark.

Although there were interactional effects between sampling dates and fractions with respect to potassium content, the various fractions showed approximately the same relationships throughout the season.

EFFECTS OF HEAVY APPLICATIONS OF POTASSIUM FERTILIZERS ON POTASSIUM CONTENT AND DISTRIBUTION

Figure 1 showed that potassium had entered the tree in considerably greater amounts under heavy applications than under normal treatment. Figure 6, *A*, shows the potassium concentration in the tree tissues on June 13 for both the potassium-fertilized and the normally treated trees. The potassium content of the leaves increased considerably more than that of either the bark or the wood.

A similar relationship existed in the absolute amounts (fig. 6, *B*), the potassium content of the leaves increasing about 100 percent as a result of the treatment. The same general tissue relationships are

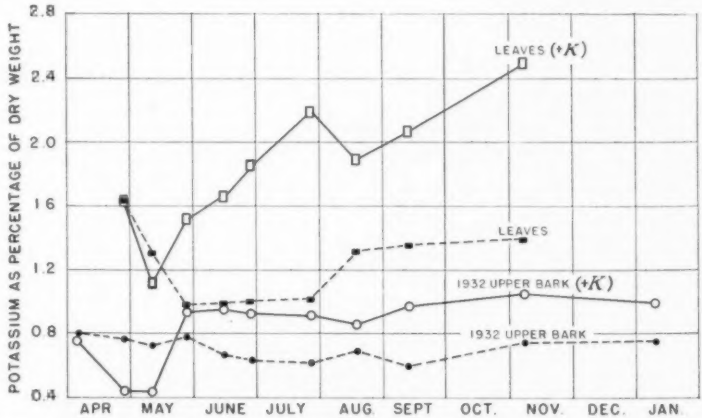

FIGURE 7.—Potassium content of the leaves and the 1932 upper bark to show the period when the increased potassium content of the potassium-fertilized trees became noticeable. +K trees treated with 1 pound potassium sulphate per tree in April; other untreated.

apparent here as in figure 5 except that the root bark was relatively low in potassium. It will be recalled that the increased movement of potassium into the roots did not take place until after June 13.

Apparently between 1 and 2 months were required for the applied potassium to enter the tree and move up to the leaves and upper twigs. Figure 7 shows the seasonal trend of the leaves and 1932 upper bark for both treatments. In both cases the treated (+K) and normal curves cross between the May 11 and May 29 sampling dates. From this period on the leaves from the trees treated with potassium fertilizer always maintained a higher concentration of potassium.

All tissues exhibited luxury consumption, but the leaves showed it to a greater degree than any other part of the tree. The leaves from the +K treatment abscissed somewhat earlier in the autumn, indicating that certain toxic, or at least detrimental, effects occurred from the heavy application of potassium.

EFFECT OF POTASSIUM APPLICATIONS ON TOTAL NITROGEN CONTENT OF TREES

Table 4 shows the total nitrogen content of the trees under both treatments throughout the season, including and excluding the leaves. Fisher's *t* comparison (*10*) made between the nitrogen

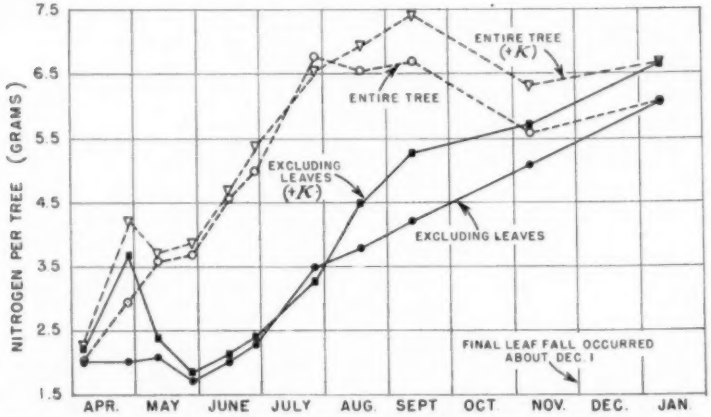

FIGURE 8.—Nitrogen content of potassium-fertilized as compared with that of normally treated trees, including and excluding leaves. +K trees treated with 1 pound potassium sulphate in April; others untreated.

TABLE 4.—*Total amount of nitrogen per tree under the two fertilizer treatments, both including and excluding the leaves, 1933*

Date sampled	Potassium-treated [1] [2]		No potassium [2]	
	Including leaves	Excluding leaves	Including leaves	Excluding leaves
	Grams	*Grams*	*Grams*	*Grams*
Apr. 8	2.3384	2.3384	2.1233	2.1233
Apr. 25	4.3412	3.7627	2.7101	2.1315
May 11	3.7784	2.4508	3.6989	2.1489
May 29	3.8362	1.9130	3.8050	1.8281
June 13	4.9359	2.1629	4.6502	2.0048
June 25			5.0258	2.2919
July 23	6.6399	3.4230	6.8996	3.4856
Aug. 16	7.1106	4.2675	6.5503	3.8200
Sept. 10	7.7134	5.2495	6.7189	4.2601
Oct. 4	6.7449	6.0522	6.4094	4.7159
Nov. 3	6.4028	5.7405	5.6270	5.0390
Jan. 8		6.7755		6.3959

[1] Treated with 1 pound potassium sulphate Apr. 1.
[2] All trees received a basic treatment of $(NH_4)_2SO_4$ and superphosphate.

content of the trees from the two treatments shows that the heavy application of potassium caused the total nitrogen content of the trees to be significantly increased. Disregarding nitrogen lost at leaf fall, the trees receiving an application of potassium absorbed 4.4371 g of nitrogen while the normally treated trees absorbed only 4.2746 g. This increase, expressed as the average of all sampling dates, is 17.7 percent. The material is presented graphically in figure 8. That the curves for the nitrogen content of the treated and untreated trees move further away from each other as the season

advances also indicates that the difference was due to treatment rather than to chance. This agrees with the work of Gildehaus (*11*) which showed that a high concentration of potassium in the culture solution resulted in a higher nitrogen content.

In an effort to determine what tissues were responsible for this phenomenon, the differences for each tissue, averaging all sampling dates, were calculated, and expressed as percentage of the nitrogen content (absolute amount) of the untreated trees. This material is presented in table 5.

TABLE 5.—*Comparison of the average nitrogen content for entire season of different fractions of trees fertilized with potassium sulphate with that of trees denied potassium*

Tissue	Treated minus untreated	Increase (+) or decrease (−)	Tissue	Treated minus untreated	Increase (+) or decrease (−)
	Grams	*Percent*		*Grams*	*Percent*
Leaves	−0.018	−0.9	Upper 1931 bark	−.001	−0.7
Upper 1933 bark	.062	35.0	Upper 1931 wood	.021	8.8
Upper 1933 wood	.053	46.9	Lower 1931 bark	.008	4.1
Lower 1933 bark	.023	10.1	Lower 1931 wood	.091	19.5
Lower 1933 wood	.180	109.7	Large root bark	.026	12.3
Upper 1932 bark	.004	3.4	Large root wood	.013	2.0
Upper 1932 wood	−.001	−6.8	Small root bark	.038	21.0
Lower 1932 bark	−.005	−4.0	Small root wood	.012	4.9
Lower 1932 wood	.016	10.4	Rootlets	−.004	−3.2

Any exact statement concerning the tissues is impossible. The current season's growth, however, showed the greatest difference, with the root bark next in order of importance. In general, except in the roots, the wood exhibited greater differences than the bark. There was essentially no difference between the leaf tissues. The importance of this relationship could hardly be conjectured, though it might designate some relationship between potassium supply and protein synthesis.

ASSOCIATION OF POTASSIUM WITH NITROGEN IN THE VARIOUS TISSUES THROUGHOUT THE SEASON

Nitrogen enters into many organic compounds in the plant, while potassium is believed to be largely in solution as inorganic salts. However, both elements are associated with living tissue, and it is of interest to compare their behavior in apple trees.

The total absorption curves of potassium and nitrogen have already been presented in figures 1 and 8 respectively. Comparing the normal treatment in both cases, the curves are found to be similar, with two exceptions, (1) although nitrogen and potassium moved into the leaves rapidly during the latter part of May, absorption of nitrogen from the soil was much slower at that time; (2) nitrogen increased from November 3 to January 8 while potassium remained rather constant This latter result seems significant, although it would require duplication before it could be deemed valid.

Figure 9, *A*, shows the percentage of the total nitrogen present in the different-aged portions of the tree and in the roots throughout the season. The data are rather similar to the corresponding potassium data shown in figure 1, *A*, the only exceptions being that a larger proportion of nitrogen resided in the roots than in the 1931 growth

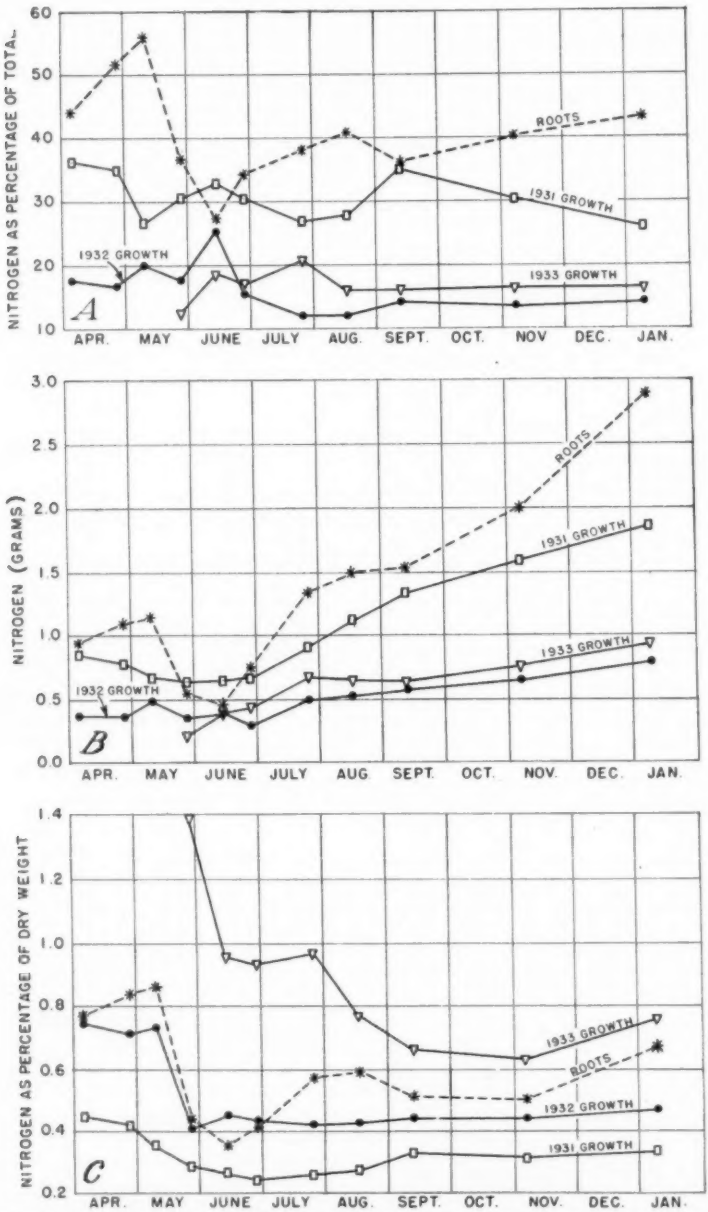

FIGURE 9.—Nitrogen content of the different-aged portions of the trees throughout the season expressed as: *A*, Percentage of the total; *B*, grams per portion; *C*, percentage of dry matter.

at the beginning of the season, and nitrogen seemed to move out of the roots and into the upper portion of the tree more rapidly during May. During the latter part of June the roots increased in nitrogen as well as in potassium and the increase continued generally throughout the remainder of the sampling period.

The data calculated on an absolute basis are shown in figure 9, *B*. Here the results closely resemble the potassium data, the only conspicuous differences being that there was a low point in the nitrogen content during June and the nitrogen content of the root fraction increased greatly after November 3. In the case of potassium, the

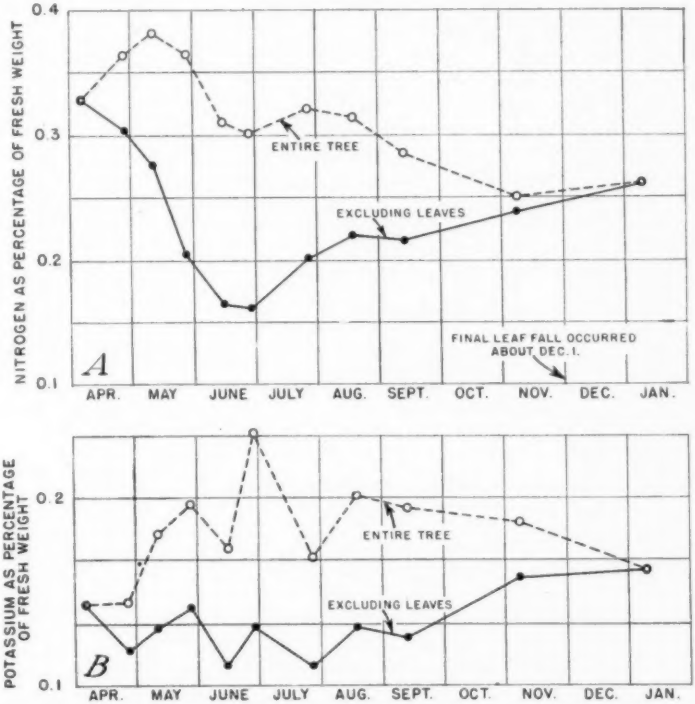

FIGURE 10.—Nitrogen (*A*) and potassium (*B*) content of the trees as a whole throughout the season, both including and excluding the leaves. Final leaf fall occurred about December 1.

increase on an absolute basis was rapid in all fractions during June and there was no increase in the roots after November 3.

The data on a percentage dry-weight basis are shown in figure 9, *C*. The curves for nitrogen and potassium again are similar, except that the roots decreased greatly in nitrogen during May and early June, and increased after November 3. Furthermore, the concentration of nitrogen in the 1932 growth dropped off during May much more rapidly than did the potassium concentration.

When the same data were calculated for the bark and wood separately, it appeared that the decrease in nitrogen on a percentage

basis in the 1933 wood was caused largely by changes in the wood rather than in the bark. In the lower portions of the tree the nitrogen changes in bark and wood were rather highly correlated.

Lincoln and Bennett (*12*) report that with pear trees the nitrogen content on a percentage fresh-weight basis remains rather constant for the entire tree throughout the season, but when the leaves are excluded from the calculation, the percentage composition drops to low values during the middle of the summer. In view of these results, it is interesting to compare similar data with apple trees, and also to see whether potassium presents a similar picture.

Figure 10 shows the percentage of fresh weight for the nitrogen and potassium content of the tree throughout the season.

In general the principle reported by Lincoln and Bennett held in the case of nitrogen (fig. 10, *A*). However, the percentage com-

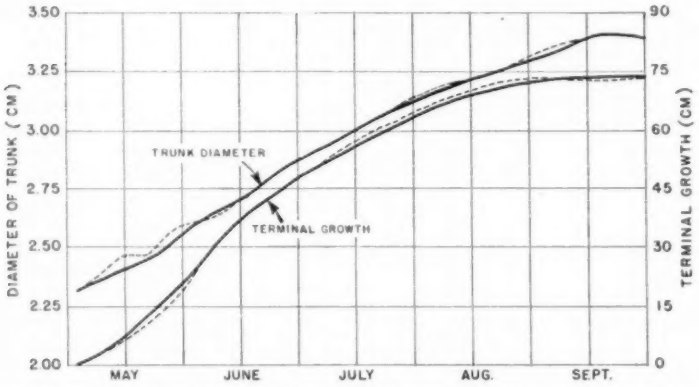

FIGURE 11.—Seasonal growth curves of trees treated with potassium sulphate as compared with those of trees denied potassium. Dotted lines indicate growth of potassium-treated trees when other than normal.

position of the tree as a whole increased during the early part of the season and decreased generally from May to November. In other words, the increase in nitrogen during April and early May was greater than the increase in fresh weight, whereas during the summer the reverse was true. During the winter, after weight increase had ceased, nitrogen absorption must be assumed to account for the increased percentage composition.

The potassium percentages did not act similarly (fig. 10, *B*); the potassium content, exclusive of that in the leaves, tended to remain somewhat more constant throughout the season, while in the tree as a whole it increased during the summer. In comparison, nitrogen moved into the leaves without being replaced in the tree proper until later in the season, while potassium was replaced nearly sufficiently in the bark and wood to make up for any potassium that moved into the leaves.

GROWTH RESPONSES

Figure 11 shows the growth curves of the trunks and terminals of the trees. It is evident that potassium fertilizer had no appreciable effect on the growth of the trees. This conformity of growth under

the two treatments is somewhat surprising since the treated trees contained so much more potassium as well as more nitrogen than the untreated ones.

DISCUSSION OF RESULTS

In the light of the findings of this study, it may be stated that the absorption of potassium by apple trees is proportional to the accumlation of dry weight. Under the high potassium treatment, potassium was absorbed at a greater proportionate rate than dry weight was accumulated. Apparently potassium was absorbed largely during the growing period, although it is certainly abundant in the soil solution during the winter. The trees apparently were able to absorb nitrogen after the first part of November, indicating that the tree is able to absorb certain nutrients even when its metabolic rate is considered to be rather low.

One of the outstanding tissue relationships noted in connection with potassium movement was the marked increase in the potassium content of the roots during the latter part of the growing season. The causal factors involved are not understood. Butler and his associates (*5*) have reported an increase in starch and sucrose in the roots toward the close of the growing season, and if potassium metabolism is similar to that of carbohydrates there is a possible utilization of additional potassium in the roots at this time, perhaps in the role of aiding translocation, as suggested by some workers. Furthermore, the same potassium might function anew in the spring when the stored carbohydrates were broken down and translocated to more active parts of the tree.

This investigation has substantiated the conclusion, already reached with other plants, that potassium is definitely associated with meristematic tissue, is very mobile, and seems to accumulate in certain tissues toward the close of the growing season.

The fact that an extremely large proportion of potassium is located in the leaves is interesting and may add to the importance of leaf-area relationships in fruit-tree responses. The fact that the leaves on the trees from the potassium-treated plots abscissed earlier in the fall may indicate that any excess of potassium is translocated to the leaves for disposal, thus partially accounting for the large luxury consumption in the leaves.

The decreasing concentration gradient of both nitrogen and potassium from the apex to the base of the tree is most logically accounted for by the decrease in the proportion of living to dead cells. The absolute-amount gradient is in the opposite direction, due to the fact that the dry weight of the fractions decreased markedly from the base to the top of the tree.

The relation of potassium to nitrogen seems to be largely a common cause association; that is, nitrogen and potassium both being essential to the life of the cell, more nitrogen and potassium occur where more living cells exist. However, the removal of nitrogen from the bark and wood of the tree as leaf formation takes place, is very marked, whereas no such movement occurs with potassium. Apparently potassium is absorbed from the soil rapidly enough to maintain a supply in the bark and wood and still supply the leaves with adequate amounts.

The increased nitrogen content of the potassium-fertilized trees cannot be adequately explained. If more growth had been produced by the fertilized trees, possibly it would account for the phenomenon. However, the fact that the greatest increase in nitrogen occurred after growth had slowed down would seem to reduce the possibility of any growth response resulting from the nitrogen content. The fact that the increased nitrogen was largely in the wood and root bark might suggest a relationship between potassium and the storage of nitrogen. Any increased supply of nitrogen in a plant, without growth correlation, would possibly result in more reserve nitrogen. Colby (*6*) showed that a potassium deficiency reduced nitrate absorption, but his finding is hardly applicable in the present case since no symptoms of potassium deficiency were observed.

The increased absorption of nitrogen subsequent to leaf fall is in keeping with the findings of Bauer (*3*) in his work with the horse-chestnut and of Combes (*7*) with the beech tree. However, other forest-tree species show other periods of nitrogen absorption. Aldrich (*1*) and Sullivan and Kraybill (*17*), working with the apple, and Schrader (*15*), with the grape, noted marked absorption of nitrogen in the fall.

Relative to the lack of growth response to the applied potassium, the fact should be stressed that if sufficient potassium accumulated in the leaves under the heavy potassium application to cause early abscission, it might have been sufficient to exercise a detrimental effect on growth and thus offset any beneficial effect of potassium fertilizers. On the other hand, the accumulation of nitrogen or potassium in one growing season might have a beneficial effect on growth the following year.

SUMMARY AND CONCLUSIONS

The absorption of potassium by young apple trees was proportional to dry-weight accumulation; it started slowly, continued at a rather rapid rate during the growing season, and then slowed down as cessation of growth approached in the fall.

The trends for the relative amounts of potassium showed that the new growth generally increased throughout the season, while the "1-year" and "2-year" tissues decreased in their relative proportions.

The amount of potassium in the roots seemed to decrease markedly during the time of rapid growth, but increased during the latter part of the season.

The absolute amount of potassium in all portions of the tree generally increased throughout the season. This was not true of the leaves, however, after abscission began.

On a percentage dry-weight basis the potassium content of the current season's growth decreased throughout the season. The roots tended to increase in potassium concentration during the last few months of growth. The 1-year growth increased in percentage of potassium during May but later decreased. The 2-year growth remained constant in its potassium concentration.

The new wood added by diameter growth corresponded closely to the current season's twig growth with respect to potassium concentration.

Young leaves contained much more potassium than old leaves. This difference may have been due either to leaching or to migration.

Generally the concentration of potassium decreased from the apex to the base of the tree, while in actual amount present the reverse was true.

The application of potassium to the soil caused an increased intake of potassium by the tree, and an increased concentration in all parts. The greatest increase was in the leaves, the next in the bark, and the lowest in the wood.

Heavy applications of potassium increased the nitrogen content of the trees.

Nitrogen and potassium were rather highly correlated in the trees. The most noticeable differences were that nitrogen was absorbed late in the season while potassium absorption stopped at leaf fall, and the nitrogen content of the tree was depleted considerably during the summer by the movement of nitrogen into the leaves. With potassium, absorption from the soil was able to maintain a uniform concentration in the wood and bark, and still maintain the content of the leaves.

There were no growth differences among the trees which could be attributed to the influence of potassium fertilizer.

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DISTRIBUTION OF TOTAL SOLUBLE SOLIDS AND CATALASE IN DIFFERENT PARTS OF JONATHAN APPLES[1]

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INTRODUCTION

The purpose of the present paper is to present the results of some studies on the variation of concentration and distribution of total soluble solids and catalase in different parts of normal and soft-scalded Jonathan apples (*Malus sylvestris* Mill.). It is not the aim or purpose to attempt any correlations or relationship with soft scald of apples, as the investigation is still in progress and the data presented herein are only preliminary in nature. The data show a rather wide variation in total soluble solids and particularly in the catalase activity between affected and sound apples and between different parts of the same apple. Other chemical fractions would probably show similar variations. That different sections of tissue, although anatomically similar, vary in certain chemical constituents and enzymic activity is emphasized.

REVIEW OF LITERATURE

A number of workers have noted wide differences in chemical constituents in different parts of horticultural fruits and in different plant materials (*7*, *8*, *11*, *12*).[2]

Lall (*11*) found that halves of the same apple varied in sugar content, although this variation was considerably less than that between different apples. Estimates of the sugar content of different parts of Bramley Seedling apples were made by Haynes and Archbold (*9*) and Archbold and Barter (*1*) with the object of determining the most satisfactory method of cutting, so far as symmetrical distribution of sugar was concerned. Haynes and Archbold show that sugar concentration increases from the stalk (stem) end toward the calyx, from the inside toward the outside, and is lower on the unblushed than on the blushed side of the fruit.

Fisher, Harley, and Brooks (*3*), working with water core of apples, found that in its early development, when the affected tissues were very localized, these parts were characterized by rapid starch conversion with a corresponding increase in soluble sugars as compared with adjacent unaffected tissues. These workers also found a very pronounced decrease in titratable acidity associated with the increase of moisture and soluble sugars in water-cored apples.

Miller (*13*) determined acetaldehyde and ethyl alcohol on duplicate samples of Jonathan apples, and found variations in the amount of these constituents in the different parts of the fruit. There was an apparent accumulation of acetaldehyde and alcohol in the peel as compared with the pulp and cores, and the concentration was much higher in the peel of fruits with soft scald than in the normal peel.

[1] Received for publication Dec. 6, 1935; issued August 1936.

[2] Reference is made by number (italic) to Literature Cited, p. 47.

METHODS

The apples for the present investigation were all of the Jonathan variety and were obtained from orchards at Hancock, Md. The fruit was picked September 12, 1935, trucked the same day to the cold-storage laboratory at the Arlington Experiment Farm, Rosslyn, Va. (near Washington, D. C.), and held under temperature conditions conducive to the development of soft scald. These conditions consisted of 6 days' delayed storage at 65° F., followed by transfer of the fruit to 32° for the remainder of the 90-day holding test. This particular study was a part of a general investigation on soft scald of apples.

The method employed for taking the pulp sample was similar to that reported by Carrick (*2*) and Harding (*5*). A cylinder of tissue was taken through the transverse center of 25 to 30 apples by means of a cork borer. From this tissue, sections representing various locations (fig. 1) were cut and composited. Because of the variation in ground color of the fruit, the skin sample was obtained by peeling very thin a narrow section around the greatest transverse diameter of the apple.

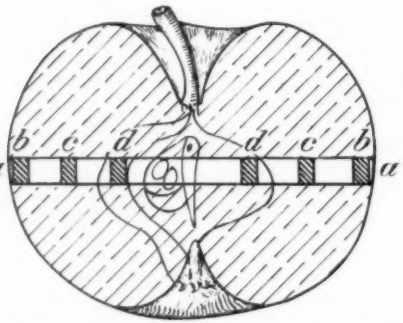

FIGURE 1.—Section of apple showing location of cylinder removed for sampling. Skin and hatched portions of cylinder represent material used for analysis: *a*, Skin; *b*, one-sixteenth inch beneath skin; *c*, halfway between skin and core; *d*, pith region.

Total soluble solids were determined by refractometer readings made on the juice extracted from each sample after grinding in a mortar and straining through a double thickness of muslin. The usual precautions for accurate refractometer determinations were taken, and corrections were made for temperature variations. Duplicate samples of juice were prepared from each lot of fruit and two refractometer readings were made on each sample. These readings in every case checked within two points in the fourth decimal place.

For catalase determinations, 4 g of tissue (skin or pulp) was weighed immediately and mixed with an equal weight of calcium carbonate and 2 ml of distilled water. An excess of calcium carbonate was used to neutralize the juice. To eliminate any differences in pH, which might possibly cause variations in catalase activity, the same amount of calcium carbonate was used in all catalase determinations. The sample was ground in a mortar until a creamy mixture was obtained. In the case of the skin sample, a small amount of clean quartz sand was added to facilitate grinding. The suspension was brought to a volume of 25 ml, with the exception of the samples of the skin and pith region, where greater dilutions were necessary in order to make readings on the measuring and leveling burettes having a capacity of 50 ml. Where the greater dilutions were made, the final catalase reading was calculated so as to be representative of the same weight of tissue.

Two samples of Komer's Antegin (catalase activator) were obtained from the Bureau of Chemistry and Soils, United States Department of Agriculture, and tested with apple tissue. When used at dilutions of 1 to 100, 1 to 500, and 1 to 1,000, there was little or no increase in catalase activity.

Qualitative tests for peroxidase determinations were made and positive results were obtained. It was difficult to evaluate the amount of the enzyme present in the skin region. The pith and regions surrounding the core indicated high peroxidase activity. The same region was also high in catalase activity. Undoubtedly peroxidase affected the results obtained for catalase. However, the manometer method for catalase determination is one of the standard methods and the error from the decomposition of hydrogen peroxide by peroxidase is believed to have been a constant error.

Standard procedure for determining catalase activity was followed, similar to that used by Haber (*4*) and Harding (*5*). Five milliliters of the skin or pulp-tissue suspension was placed in one arm of the catalase tube and 5 ml of hydrogen peroxide was placed in the other. The tube was suspended in a water bath held at a uniform temperature of 70° F., and the solutions in the tube were allowed to reach the temperature of the water bath. Figure 2 shows details of the apparatus. The samples were shaken 15 minutes at a uniform rate of 120 oscillations per minute, after which the amount of evolved oxygen was measured. Every precaution was taken to obtain a uniform sample and a uniform suspension of the macerated tissue. The duplicate samples, or samples from the same part, whether skin or pulp, checked within 0.3 ml of evolved oxygen.

DISTRIBUTION OF TOTAL SOLUBLE SOLIDS

Table 1 shows the amount of total soluble solids as they were found distributed throughout various parts of the apple. It will be noted that the skin tissue contained the largest amount, and that the concentration decreased toward the core region. This is in harmony with the findings of Haynes and Archbold (*9*), who concluded that sugar concentration decreased from the outside toward the inside of the fruit. It might be noted also that samples from normal Jonathan apples had slightly greater amounts of total soluble solids than did comparable samples from apples showing soft scald (table 1).

TABLE 1.—*Concentration and distribution of total soluble solids and catalase activity in normal and soft-scalded Jonathan apples* [1]

Sampling region	Total soluble solids		Catalase activity	
	Normal apples	Soft-scalded apples	Normal apples	Soft-scalded apples
	Percent	*Percent*	*Milliliters* [2]	*Milliliters* [2]
Skin only	15.96	[3] 15.47	95.00	[3] 4.00
1/16 inch beneath skin	13.23	[4] 12.47	11.60	[4] 4.10
Halfway between skin and core	13.03	[5] 12.97	19.00	[5] 27.60
Pith [6]	12.72	[5] 12.42	34.50	[5] 45.00

[1] 4 pairs of samples, or 8 altogether, consisting of from 25 to 30 apples each, were used. Each of the 8 samples was tested in duplicate, and the final results shown are averages from the 4 analyses of each pair of samples.
[2] Milliliters of oxygen evolved at the end of 15 minutes.
[3] Skin scalded.
[4] Tissue brown.
[5] Tissue normal in appearance.
[6] Kraus (*10*) designates the region between the primary vascular bundles and core as the pith.

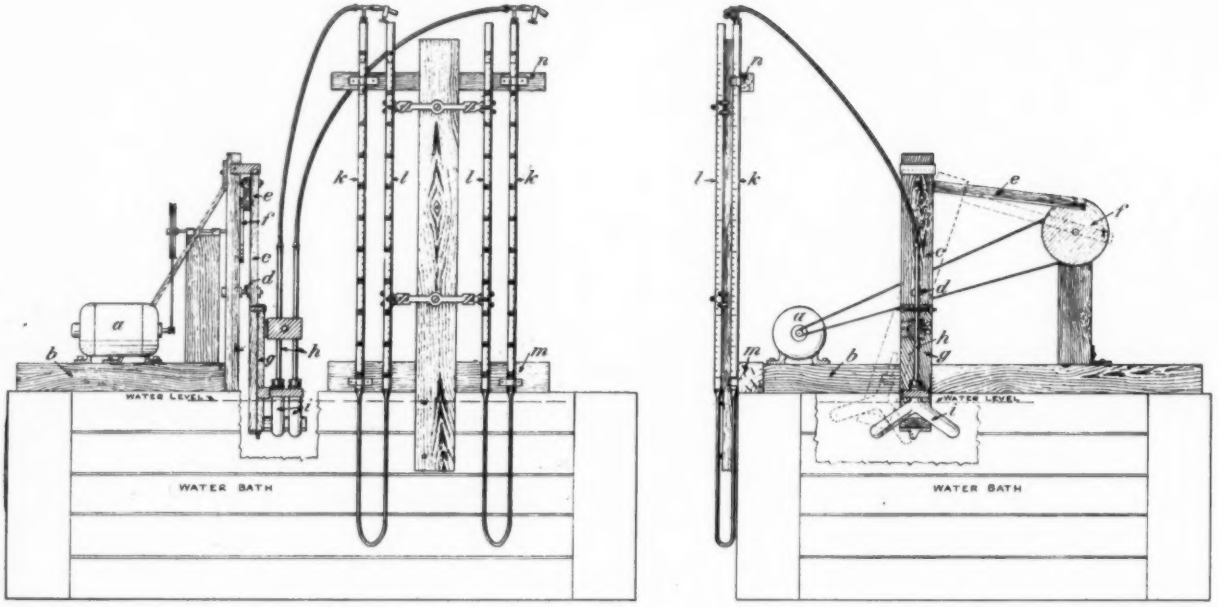

FIGURE 2.—Apparatus for duplicate catalase determinations: *A*, Front view; *B*, side view. *a*, Motor; *b*, platform over water bath; *c*, shaker arm; *d*, pivot for shaker arm; *e*, connecting rod; *f*, driving disk for connecting rod; *g*, catalase-tube holder; *h*, connecting glass tube; *i*, catalase tube; *k*, stationary burette; *l*, leveling burette; *m*, cleat; *n*, crosspiece.

DISTRIBUTION OF CATALASE

The data on the distribution of catalase activity are also presented in table 1 for purposes of convenience and without intent to imply any correlation between them and the data for total soluble solids.

In normal apples, catalase activity was highest in the skin and least in the region immediately beneath the skin, increasing toward the core (table 1). In apples showing soft scald, the least catalase activity was noted in the diseased portion comprising the skin and the brown tissue immediately beneath. It seems quite possible that in peeling the scalded skin a small amount of normal skin might have been included in the sample. This would account for the reading of 4 ml of oxygen evolved, since the brown tissue beneath the scalded skin showed only 0.1 ml of evolved oxygen (table 1).

Aside from the skin tissue, the regions of highest catalase activity were the pith and the parenchyma tissue halfway between core and skin (table 1). In these two regions catalase activity was significantly higher in soft-scalded apples than in corresponding samples from normal apples. The writer (*6*) has observed that high catalase activity was registered just prior to the appearance of soggy breakdown in Grimes Golden apples.

SUMMARY

Specific differences in total soluble solids and catalase activity were found in different parts of Jonathan apples.

Soluble solids were consistently lower in apples affected with soft scald than in those not affected, but in both cases the greatest concentration was found in the skin, with a gradual decrease toward the pith.

In normal apples, catalase activity was highest in the skin and least in the region immediately beneath the skin. In apples showing soft scald, catalase activity was highest in the pith region and lowest in the diseased portion comprising the skin and the brown tissue immediately beneath it.

The results of this investigation suggest that preliminary work to determine localized differences in different parts of an apple should precede chemical or physiological studies in which the fruit is customarily analyzed as a whole. This procedure might lead to a modification of methods of sampling and give quite different and more significant results.

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DISTRIBUTION OF ACETALDEHYDE AND ALCOHOL IN THE APPLE FRUIT[1]

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INTRODUCTION

In the course of an investigation, which is still in progress, concerning the role of acetaldehyde in the metabolism of stored fruit, several interesting observations were made regarding the distribution of acetaldehyde in the apple fruit. Studies of the distribution of soluble solids and catalase activity were conducted on the same lot of fruit by Harding (*5*).[2] The present paper reports the data concerning the distribution of acetaldehyde and alcohol in the fruit.

REVIEW OF LITERATURE

It has long been known that acetaldehyde and alcohol may appear as products of metabolism in many higher plants stored under anaerobic conditions. Thomas (*10*) has reviewed the earlier work rather thoroughly, and reference is made here only to recent literature that pertains to the distribution of these substances in the fruit. Power and Chesnut (*9*), in their work on the odorous constituents of apples, stated that these substances appeared to be contained chiefly in the outer skin or rind of the fruit, and their analyses of the apple parings indicated the presence of several different esters as well as acetaldehyde.

Gerhardt[3] has shown that the peel of Jonathan apples is higher in acetaldehyde than the pulp of either normal apples or of those showing soft scald; furthermore, that the acetaldehyde content of peel from the diseased apples is higher than that of normal apples.

Kidd and West (*7*) exposed apples to ethyl alcohol vapor and analyzed the cores and "inner" and "outer" sections of the pulp for acetaldehyde and alcohol. They reported that while the alcohol is being taken up the largest amount of ethyl alcohol is found in the cores and that while the alcohol is escaping the reverse is true. The aldehyde content during alcohol absorption was highest in the "outers", but during alcohol escape there was little difference in the acetaldehyde content of these three portions of the apple.

Fisher, Harley, and Brooks (*3*) reported that appreciable quantities of ethyl alcohol are present in the water-cored area of apple fruits and that under certain conditions relatively large quantities may be found.

[1] Received for publication Dec. 21, 1935, issued August 1936.

[2] Reference is made by number (italic) to Literature Cited, p. 55.

[3] GERHARDT, F. Unpublished report. 1934.

MATERIAL AND METHODS

The Jonathan apples used in these experiments were obtained from the American Fruit Growers' orchard at Hancock, Md. They were picked September 11, 1934, delayed 6 to 10 days at 18° C., and then stored at 0° until December 11, 1934, when they were analyzed.

The Grimes Golden apples used for the study of soggy break-down were also grown at Hancock and received the same storage treatment except that they were held in storage till February 11, 1935. The Grimes Golden apples used in the experiments on freezing and those used in comparing the different methods for preparing pulp samples were grown at Vienna, Va., about 10 miles from Washington, D. C. These apples were harvested October 9, 1935, and were subjected to the experimental tests immediately after harvesting.

Thirty Jonathan apples were employed for each sample. The apples were separated into three parts—peel, pulp, and cores. The peel was removed by a mechanical peeler and the cores were cut out with a 2-cm cork borer. The peeled and cored apples were then cut into slices small enough to slip through the neck of an 800-ml Kjeldahl flask. This constituted the pulp sample.

One-hundred-gram duplicate samples were analyzed for acetaldehyde by the method described by Harley and Fisher (*6*). This consists of steam-distilling the samples for 90 minutes and collecting the distillate in a standard solution of sodium bisulphite. Inasmuch as this is a rather delicate determination, extreme precautions were observed in order to reduce the chances of error. The solutions of iodine and sodium bisulphite were standardized each day and stored in the refrigerator when not in use. The sodium bisulphite was delivered from a burette stoppered with a soda-lime tube and was not added to the receiving flasks until they had been cooled in the ice bath and were ready for immediate connection with the condensers. Instead of titrating the bisulphite with iodine, a known quantity of iodine solution was added to the distillate and this was titrated "back" with sodium thiosulphate. With these precautions, in addition to those described by Harley and Fisher, duplicate distillations were obtained which checked within 0.1 or 0.2 ml of N/10 $Na_2S_2O_3$.

Alcohol determinations were made by the method described by Thomas (*10*). This consists of removing the acetaldehyde and oxidizing the alcohol to acetic acid.

RESULTS OF ANALYSES

JONATHAN APPLES

Table 1 shows the quantities of acetaldehyde and ethyl alcohol found in the different parts of Jonathan apples. Both sound fruit and similar apples affected with soft scald were studied. There was an apparent accumulation of acetaldehyde and alcohol in the peel as compared with the pulp and cores, and the concentration was much higher in the diseased than in the normal peel.

TABLE 1.—*Acetaldehyde and ethyl alcohol content of peel, pulp, and cores of Jonathan apples*

Tissue analyzed	Acetaldehyde in—			Alcohol in—		
	Peel	Pulp	Cores	Peel	Pulp	Cores
	Mg [1]	*Mg* [1]	*Mg* [1]	*Mg* [1]	*Mg* [1]	*Mg* [1]
Normal	1.0	0.0	0.3	28.8	17.0	11.6
Soft scald	4.1	.5	.5	36.0	11.6	12.6

[1] Per 100 g of fresh tissue.

An additional lot of Jonathan apples was analyzed in which the pulp was separated into inner and outer portions. In this lot (table 2) also the peel showed the highest concentration of acetaldehyde. Each portion of the diseased apples showed a higher concentration than did the corresponding portion of the normal apples. The inner pulp was usually higher in acetaldehyde than the outer pulp.

An attempt was made to analyze the cuticle of the apples after removal by the method recommended by Markley and Sando (*8*), but it was impossible to prepare the material for analysis without introducing errors. Instead, the apples were immersed quickly in ether, rinsed in water, and dried. The peel was then analyzed for acetaldehyde and total ether extract in order to ascertain whether the removal of the ether-soluble fraction of the cuticle removed much of the acetaldehyde. From table 3 it will be noted that the rinsing in ether removed 16.3 percent of this ether-soluble fraction and 36.7 percent of the acetaldehyde.

GRIMES GOLDEN APPLES

The studies were later extended to include Grimes Golden apples showing soggy break-down. This disease is characterized by a browning of the pulp, whereas soft scald is limited to the peel and a small portion of the pulp directly beneath it. In these analyses (table 4) the peel again showed higher acetaldehyde than the pulp or cores and the peel from the diseased apples showed higher acetaldehyde content than that from the normal ones. The aldehyde content of the cores was higher in the diseased than in normal apples; the reverse was true of the pulp.

TABLE 2.—*Acetaldehyde content of peel, outer pulp, inner pulp, and cores of Jonathan apples*

Tissue analyzed	Peel	Outer pulp	Inner pulp	Cores
	Mg [1]	*Mg* [1]	*Mg* [1]	*Mg* [1]
Normal	2.0	0.3	0.65	0.45
Soft scald	6.0	1.7	2.3	.55

[1] Per 100 g of fresh tissue.

TABLE 3.—*Acetaldehyde and ether extract removed from peel by immersing Jonathan apples in ether*

Tissue analyzed	Acetaldehyde	Ether extract
	Mg [1]	*Mg* [1]
Peel	4.9	0.190
Peel washed with ether	3.1	.159
	Percent	*Percent*
Loss	36.7	16.3

[1] Per 100 g of fresh tissue.

TABLE 4.—*Acetaldehyde content of peel, pulp, and cores of Grimes Golden apples*

Tissue analyzed	Peel	Pulp	Cores
	Mg [1]	*Mg* [1]	*Mg* [1]
Normal	6.2	2.8	0.0
Soggy break-down	9.1	2.4	1.9

[1] Per 100 g of fresh tissue.

It was found that comparable duplicate results could not be obtained unless the analyses were made simultaneously. Fidler (*2*) states that sliced apples exposed to air for a half hour may lose significant amounts of acetaldehyde. In the present work the discrepancies were apparently due to accumulation rather than to loss of acetaldehyde. Samples of peel, pulp, and cores of Grimes Golden apples were weighed, transferred to 800-ml Kjeldahl flasks, stoppered, and held in the refrigerator. Acetaldehyde determinations made at the end of 1, 2, and 4 hours showed definite increases during this time (table 5). At the end of 4 hours the acetaldehyde content of the peel and pulp had approximately doubled; that in the cores had quadrupled.

TABLE 5.—*Accumulation of acetaldehyde in cut apple tissue analyzed immediately and after storage for 1, 2, and 4 hours in a flask in the refrigerator*

Time of sampling	Peel	Pulp	Cores
	Mg [1]	*Mg* [1]	*Mg* [1]
Immediately	6.8	1.1	0.9
After 1 hour		1.8	
After 2 hours	13.1	1.6	
After 4 hours	13.4	2.6	3.7

[1] Per 100 g of fresh tissue.

It has been suggested that the high acetaldehyde content of the peel in these experiments may be traceable to its rapid formation in the cells injured through peeling the apples, rather than to its adsorption by the peel. A further study of this phase was made by comparing finely sliced pulp samples with those of the peel. Thirty Grimes Golden apples were treated with carbon dioxide to increase the acetaldehyde content. The peel was removed by the mechanical peeler, and the apples were then replaced on the machine and strips of pulp were removed in the same manner as the peel. The results of the acetaldehyde determinations appear in the first line in table 6.

TABLE 6.—*Content and distribution of acetaldehyde in Grimes Golden apples under various conditions*

Treatment	Total acetaldehyde content	Acetaldehyde distribution			
		Peel	Pulp		
			Thin sections	Sliced	Finely ground
	Mg [1]	*Mg* [1]	*Mg* [1]	*Mg* [1]	*Mg* [1]
Treated with CO_2		2.2	1.5		
Normal		.6		0.0	0.4
Do	0.1				
Frozen [2]	.1				
Normal [3]	.3				
Frozen [4]	1.1				

[1] Per 100 g of fresh tissue.
[2] 18 hours in freezing room; 4 hours in laboratory.
[3] Held for 24 hours in the laboratory before being analyzed.
[4] 18 hours in freezing room; 24 hours in laboratory.

This experiment was repeated with normal Grimes Golden apples. In this instance the pulp was prepared in two ways. After the apples had been peeled and cored, one half of each of the 30 apples was cut in slices small enough to pass through the neck of an 800-ml Kjeldahl flask; the other half was ground in a food chopper. It is probable that the high acetaldehyde content of the peel, as shown in table 6, may be due both to the large number of cells injured in the peeling process and to the tendency of the acetaldehyde to accumulate in the peel.

Another question that arises is whether apple cells injured by freezing produce acetaldehyde in abnormal quantities. This was investigated in the following manner. Thirty Grimes Golden apples were held in the freezing room for 18 hours. The temperature of the room was about −8° C. The temperature of the apples when removed was −3.5° at the core. They were definitely frozen but not permanently injured. When the apples were thawed the flavor was not quite so good as that of the controls, but they would no doubt have been readily accepted by the average consumer. They were held in the laboratory for 4 hours and then analyzed for acetaldehyde. Samples consisted of the entire edible portion of the apple. Controls were held in the cold-storage room at 0° during the time the others were being frozen and were then transferred to the laboratory along with the frozen lot.

This experiment was repeated with the modification that the apples were held in the laboratory 24 hours before analyzing for acetaldehyde. It will be noted (table 6) that in the first experiment the frozen apples produced no more acetaldehyde than did the normal apples. In the second experiment, however, when the apples were held in the laboratory 24 hours following freezing, the acetaldehyde content was more than three times as great in the frozen as in the normal apples.

DISCUSSION

Other investigators have reported that the outer portion of the apple fruit is more active physiologically than the region near the core. Fellers, Isham, and Smith (*1*) have shown that the vitamin C

content of Baldwin and McIntosh apples is concentrated in the epidermis and fleshy cortex. In the Baldwin variety the epidermis was about 4 times as active in vitamin C as the flesh near it, and 6 to 10 times as active as the flesh in the pulpy area near the core. Harding (*5*) found that in normal Jonathan apples the skin showed both higher catalase activity and higher soluble solids than the pulp at different depths between the peel and core.

In earlier stages of the present investigation it appeared that acetaldehyde was produced most extensively in that portion of the apple that is removed by a mechanical peeler. Subsequent experiments, however, suggested that the amount of acetaldehyde formed may depend upon the number of cells ruptured. It is also possible that the acetaldehyde accumulates in the cuticle of the apple as suggested by Gane (*4*). It is significant that the peel of apples affected with either soft scald or soggy break-down always showed higher acetaldehyde content than that of normal apples, and this was true whether the disease was manifested in the peel or in the pulp of the apples. It is apparent, then, that whether the apple is producing acetaldehyde as a result of physiological disorder or of mechanical injury the acetaldehyde tends to accumulate in the peel.

SUMMARY

The acetaldehyde content and the alcohol content of Jonathan apples have been shown to be higher in the peel than in the pulp or cores, and higher in the peel of soft-scalded Jonathan apples than in that of normal apples.

Rinsing Jonathan apples in ether removed 16.3 percent of the total ether extract of the peel. As a result of this process the peel lost also 36.7 percent of its original acetaldehyde content.

The acetaldehyde content of normal Grimes Golden apples is higher in the peel than in the pulp or cores. Acetaldehyde content is higher in the peel of Grimes Golden apples showing soggy break-down than in that of normal apples.

Samples of peel, pulp, and cores prepared for analyses but held in stoppered flasks in the refrigerator 4 hours accumulated from two to four times the original amount of acetaldehyde.

Finely ground pulp of normal Grimes Golden apples yielded more acetaldehyde than coarsely sliced pulp but not so much as the peel.

Frozen Grimes Golden apples when sampled 4 hours after freezing showed no more acetaldehyde than normal apples. When held 24 hours in the laboratory following freezing the acetaldehyde content was more than three times as great in the frozen apples as in the normal apples.

It appears that high acetaldehyde content of apple peel may be due in part to its production by cells ruptured in the paring process, but there is also a tendency for the acetaldehyde to accumulate in peel affected either by mechanical injury to the cells or by abnormal physiological conditions of the fruit.

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THE ROLE OF THE CAPILLARY POTENTIAL IN THE DYNAMICS OF SOIL MOISTURE[1]

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For the movement of water horizontally it is natural to assume that the greater the pressure difference between neighboring points the greater will be the velocity. Darcy discovered a relation which is expressed in the following simple form:

$$v=\frac{k(p_1-p_2)}{l} \tag{1}$$

where v is the velocity of water flowing through a horizontal pipe containing sand, p_1 the pressure at one end and p_2 the pressure at the other end, l the length of the tube, and k a constant of proportionality. This law has been observed by several investigators, but its far-reaching significance has not been widely known or understood.

It is apparent that for vertical flow the equation is inadequate. The factor $\frac{p_1-p_2}{l}$ represents in this special case a vector quantity, which has been called the gradient of the pressure p. It is the force per unit volume due to variation in pressure from point to point. It may be expressed by the concise symbol ∇p. Dividing ∇p by the density ρ of the fluid reduces it to a force per unit mass. Conservative forces (forces which do not arise from friction) are frequently expressed as a matter of convenience as the gradients of potential functions. Thus, the gravitational field intensity g is set equal to the negative gradient of the gravitational potential function ϕ

$$g=-\nabla\phi \tag{2}$$

In the case of horizontal flow the direct effect of gravity is balanced out in such a way as to have no influence on the motion, and for this reason does not appear in equation (1). For the general case it is necessary to modify the equation thus

$$v=-k\rho((1/\rho)\nabla p+\nabla\phi) \tag{3}$$

For velocities which are reasonably small this equation represents a successful dynamical background for the movement of water in saturated soils.

Conciseness may be gained by introducing a new symbol for the first term in the parenthesis of the right-hand side of equation (3). If we take into account the slight compressibility of water, and

[1] Received for publication Feb. 15, 1936; issued August 1936. Contribution from the department of physics, Utah Agricultural Experiment Station.

therefore the slight variation in ρ, a function ψ of the position coordinates, x, y, z, satisfying the following vector equation

$$\nabla\psi=(1/\rho)\nabla p \tag{4}$$

may be found, provided only that ρ is a function of p. Except for an arbitrary integration constant this function differs only slightly from the ratio p/ρ, and it is to this function that the name capillary potential has been given.

If we introduce the symbol Φ, defined thus

$$\Phi=\psi+\phi \tag{5}$$

it is possible to write the generalized Darcy equation in the concise form

$$v=-k\rho\nabla\Phi \tag{6}$$

Whether or not the concise symbolism is to be desired, it still remains a fact that the physical quantity ψ, which we call the capillary potential, plays a significant role in this important basic equation.

This method of attack has suppressed the fact that the actual region occupied by water in the soil is bounded by a much distorted and complicated solid-water interface. For unsaturated soils a new bounding air-water interface appears, and if we confine our attention to the water region thus bounded we are still confronted with the physical concepts of pressure, density, capillary potential, etc., and they have the same meaning except that the pressure is throughout the region less than atmospheric and it might more appropriately be designated as capillary tension. The term "capillary potential" was introduced first by Buckingham[2] in a discussion of moisture phenomena in unsaturated soils, and in the literature published at the Utah Agricultural Experiment Station its connection with unsaturated soils has been stressed. Although defined with reference to this water phase it also will be construed in a macroscopic sense as a point-characteristic of the soil.

To measure it a porous cup filled with water is brought into close contact with the soil and the tension in the soil water is transmitted to the water in the cup and thence to an ordinary manometer. For soils sufficiently dry to have a "tension" as great as an atmosphere this method is inadequate.

It is interesting to observe, however, that there is a close connection between the capillary potential and the corresponding vapor pressure. If water is permitted to ascend a column of soil from a free-water surface it is evident that at each point above the free water when equilibrium is established a definite moisture percentage, a definite capillary potential, and a definite vapor pressure will be established. The capillary tension at a height h from free water would be ρgh, whereas the vapor pressure π would decrease exponentially from a value π_0, thus,

$$\pi=\pi_0 e^{-kh} \tag{7}$$

or, in another form,

$$h=-\frac{\log_e(\pi/\pi_0)}{k} \tag{8}$$

[2] BUCKINGHAM, E. STUDIES ON THE MOVEMENT OF SOIL MOISTURE. U. S. Dept. Agr. Bur. Soils Bull. 38, 61 pp., illus. 1907.

leading to the relation

$$\psi = -\frac{g \log_e (\pi/\pi_0)}{k} \tag{9}$$

It seems evident a priori that the constant factor $k\rho$ can no longer completely represent the so-called transmission constant, but that some as yet undetermined function $f_1(\rho')$ of the variable moisture content ρ' of the soil, involving parameters characteristic of the soil, should be substituted. Furthermore, although, as stated, there is no essential change in the connection between the capillary potential and the pressure and density of the water, its actual magnitude is influenced by the amount of moisture in the soil. It will be appropriate, therefore, to introduce for the capillary potential another function $f_2(\rho')$, this function also containing characteristics oil parameters. The Darcy law may thus be generalized to apply to unsaturated soils as follows:

$$v = f_1(\rho')\nabla(f_2(\rho') + \phi) \tag{10}$$

In addition to the Darcy velocity law we have another differential equation which comes directly from the law of conservation of matter and which is called the equation of continuity. To express this equation in concise form a new symbol $\nabla\cdot$, known as the divergence operator, is introduced, thus,

$$\nabla\cdot(\rho' v) = -\partial\rho'/\partial t) \tag{11}$$

This equation means that the divergence of the flux density is equal to the negative time rate of change of the density. Eliminating v from this equation by means of equation (10), we obtain

$$\rho' f_1 \nabla^2 f_2 + (\nabla(\rho' f_1))\cdot(\nabla f_2 + \nabla\phi) = -\partial\rho'/\partial t \tag{12}$$

A quantity $\nabla^2\phi$, which vanishes on account of the constancy of $\nabla\phi$, has been omitted from the equation. The dot between the parentheses of the second term indicates that the scalar product of the two vectors is to be taken.

New experimental work is required to determine the functions f_1 and f_2. On the basis of the original Darcy law they are known of course for saturated soils, and for the case of steady flow equation (12) reduces to the well-known Laplace equation and it forms the basis for the solution of practical problems concerning the movement of ground water.

Tentative approximations also have been presented for the case of unsaturated soils. The physics laboratory at the Utah station has made use of the following simple substitutions

$$f_1 = k\rho' \tag{13}$$

and

$$f_2 = c/\rho' + b \tag{14}$$

where k, c, and b are empirical constants. Experimental results published heretofore indicate that these represent good first approximations.

Much has been written in texts on applied mathematics concerning the solution of the Laplace equation for various boundary conditions.

A more detailed discussion of the ground-water problem, together with a special application of equation (12) subject to the approximations (13) and (14), has been presented recently.[3]

Various soil characteristics have been defined all of which no doubt serve some useful purpose. It is to be observed, however, that such characteristics as the transmission factor and the capillary potential enter naturally into the basic analysis of the fundamental dynamical problem of the movement of soil moisture, and it is this problem that is of primary concern to irrigation and drainage engineers. Their application to the so-called capillary fringe has not been extensive. It is nevertheless significant that capillary phenomena have constituted a nucleus about which has centered much discussion in researches in soil physics since its beginning, and it can scarcely be claimed that the problem has been satisfactorily solved without this or some similar approach from the standpoint of classical dynamics.

[3] GARDNER, W., COLLIER, T. R., and FARR, D. GROUND-WATER, PART 1. FUNDAMENTAL PRINCIPLES GOVERNING ITS PHYSICAL CONTROL. Utah Agr. Expt. Sta. Bull. 252, 40 pp., illus. 1934.

THE EFFECT OF FERTILIZERS AND LIME UPON THE ELECTRODIALYZABLE AND EXCHANGEABLE POTASH OF CROPPED SOIL.[1]

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INTRODUCTION

The fertility plots maintained at the experimental farm of the Delaware Agriculture Experiment Station offered an opportunity for investigating the effects of fertilizers, manure, and lime upon the electrodialyzable and exchangeable potash of plot soils. The plots were established in 1908 and have been subjected to a definite cropping system and fertilizer program.

REVIEW OF LITERATURE

Humfeld (*8*)[2] found that fertilizer treatments continued over a term of years have a measurable effect upon the kind and amounts of bases and acids extracted from the soil by electrodialysis.

Holmes (*7*) and Schollenberger and Dreibelbis (*14*) found that applications of manure resulted in a slight increase in exchangeable potassium, but Merkle (*13*) noted no increase.

Volk (*17*) and Bartholomew and Janssen (*3*) obtained evidence of the fixation of added potash in nonreplaceable form.

Snider (*16*) found that replaceable potassium was increased by the continued use of potash salts in Illinois plots, and observed a tendency toward lower replaceable potassium values on the heavily limed soils as compared with the lightly limed. Brewer and Rankin (*4*) also found that liming decreased the amount of potassium removed by electrodialysis and the Neubauer method. Abel and Magistad (*1*), however, found that on cropped and fallow pineapple soils liming increased the replaceable potassium, and the crops removed more potassium from the limed soils. In soils deficient in potash the crops removed more potassium than was present in replaceable form, yet in some cases the amount of replaceable potassium was greater at the end of the experiment than at the beginning. Their results are in agreement with those of Hoagland and Martin (*5*, *6*).

MacIntire and his coworkers (*12*, *p. 509*) concluded from the results of lysimeter studies that:

> As a whole, the results established the fact that the liming of rock-derived soils under humid conditions will depress the hydrolytic disintegration of both the original potassium complex and that formed by the fixation of soluble added potassium salts, and further that the protective, or buffering, effect becomes more pronounced with increased concentration of the bicarbonates of calcium and magnesium.

[1] Received for publication Jan. 24, 1936; issued August 1936.

[2] Reference is made by number (italic) to Literature Cited, p. 65.

Jenny and Shade (*9*), however, found that potassium was liberated by the calcium ion in the presence of Cl, SO_4, CO_3, HCO_3, OH, and PO_4 ions. Their investigation did not include a study of the conversion of nonreplaceable to available potassium.

EXPERIMENTAL METHODS

The plots from which the samples [3] were taken are designated as "block B" of the fertility plots at the experimental farm. The soil is classified as Sassafras silt loam.

The treatments to which the plots were subjected over a period of 24 years are indicated in table 1.

TABLE 1.—*Treatment of the block B fertility plots, 1908–32*

[Quantities applied are per acre]

Plot no.[1]	Muriate of potash	Sodium nitrate	Dried blood	Superphosphate	Farm manure
	Pounds	*Pounds*	*Pounds*	*Pounds*	*Tons*
4	1,335				
7	1,335			3,900	
8	1,335	1,675			
9	1,335	1,599	75	3,900	
10	2,550	3,150	100	7,300	
14				750	65
15					130

[1] Nos. 6, 11, and 16 were check plots.

Each plot (one-tenth of an acre) was divided into two sections, one of which received lime as follows: 1908 and 1916 at the rate of 2,000 pounds per acre; 1912, 1920, and 1924 at the rate of 1,500 pounds per acre. The last application of lime was made in 1924. Both the limed and unlimed sections received the same fertilizer and manure treatments.

The cropping system employed from 1908 to 1912 was corn, oats, wheat, and timothy and clover; since 1912 it has been corn, soybeans, wheat, and timothy and clover.

Samples were taken in the late summer of 1932. Twenty cores (1⅝ by 6⅔ inches) were taken from each section (lime and unlimed) of each plot with the utmost care to insure samples as representative as possible. The soil was thoroughly mixed, air-dried, and again quartered.

One hundred grams of air-dried soil was electrodialized in a three-compartment Mattson cell. Parchment membranes and perforated sheet-platinum electrodes were employed. With this arrangement the electrodes were approximately 2 cm apart. Current was supplied by a motor generator and applied at 110 volts. The soil was subjected to electrodialysis for 1 hour, the dialyzates removed, and electrodialysis continued for three 8-hour periods, making a total of 25 hours for each sample. Potash was determined in the dialyzate.

One hundred grams of air-dried soil was leached with normal ammonium acetate according to the method of Schollenberger and Dreibelbis (*15*) and potash was determined in the leachate.

[3] The soil samples and data in table 1 were furnished by Dr. H. C. Harris, assistant agronomist of the station.

Hydrogen-ion determinations were made on the soils after they had stood for 3 days, with occasional shaking, in 2.5 times their weight of water. The MacInnes-Dole type glass electrode with vacuum-tube potentiometer was employed.

RESULTS

The course of the current during electrodialysis was remarkably uniform throughout the series, rising to a maximum of approximately 300 milliamperes within a few minutes and decreasing gradually as dialysis progressed until it became practically constant at 30 milliamperes.

The results for dialyzable, exchangeable, and total potash and the pH values for the plot soils are presented in table 2.

Without exception, more potash was removed by electrodialysis from the limed soils than from the unlimed. With the exception of plots 8 and 16, in which the values for limed and unlimed sections are identical, this holds true for exchangeable potash. While exchangeable and dialyzable potash are of the same order, exchange potash is of lesser magnitude. Applications of potash are reflected to a greater degree by electrodialysis than by the exchange method. Both exchangeable and dialyzable potash are at a maximum in plot 15, which received manure only. The total potash varies between 39.7 milliequivalents (plot 4) and 35.3 milliequivalents (plots 14 and 15) in the limed series and between 36.1 milliequivalents (plot 16 check) and 32.9 milliequivalents (plot 11 check) in the unlimed series. In general, however, the potash content is quite uniform throughout each series with that of the limed soil exceeding in every case that of the corresponding unlimed soil.

The pH values are rather uniform throughout the respective series (limed and unlimed) with the lowest for plot 4 (KCl not limed) and the highest for the check plots 6 and 11 (limed).

TABLE 2.—*Effect of fertilizers, manure, and lime upon the electrodialyzable and exchangeable potash of sassafras silt loam*

[Quantities per 100 g of soil dried at 110° C. for 18 hours]

Plot no.	Treatment	Limed	pH	K_2O		
				Total	Electrodialyzable	Exchangeable
				Milliequivalents	*Milliequivalents*	*Milliequivalents*
4	K	Yes	6.72	39.7	0.125	0.109
		No	5.28	35.2	.111	.100
6	Check	Yes	6.92	36.1	.111	.090
		No	5.43	35.9	.075	.080
7	PK	Yes	6.87	36.7	.131	.092
		No	5.51	35.5	.102	.087
8	NK	Yes	6.71	36.0	.142	.098
		No	5.68	34.4	.126	.098
9	NPK	Yes	6.85	35.9	.146	.093
		No	5.56	34.7	.127	.070
10	2 (NPK)	Yes	6.63	35.5	.155	.092
		No	5.59	34.5	.135	.081
11	Check	Yes	6.93	36.6	.110	.098
		No	5.42	32.9	.096	.063
14	P and manure	Yes	6.80	35.3	.132	.084
		No	5.47	33.8	.109	.082
15	2 (manure)	Yes	6.64	35.3	.165	.124
		No	5.53	35.2	.129	.115
16	Check	Yes	6.70	36.3	.122	.083
		No	5.44	36.1	.106	.083

DISCUSSION

While there is evidence that applications of potash fertilizers have increased the amount of exchangeable and electrodialyzable potassium, the most consistent effects have been brought about by liming.

If it be assumed that exchangeable or electrodialyzable potassium is a reliable index to availability, the logical conclusion to be drawn from the results is that liming has increased the availability of this element. This conclusion is obviously unjustified in view of the fact that, without exception, the total potash of the limed soils exceeds that of the unlimed, and the yields from the limed soils were generally greater. It has also been shown (*6*) that a soil with a comparatively low value for replaceable potassium may still supply sufficient potassium for high yields over a considerable period of time, whereas another soil of similar replaceable potassium value may be very deficient in potassium-supplying power.

Several investigators (*2*, *10*, *11*) have found that liming decreases the amount of potassium taken up by the crop, yet Lipman, Blair, and Prince (*10*) found that the total potash of the limed soils was, in general, lower than that of the corresponding unlimed soils. The differences could not be accounted for by the increased yields on the limed soils, but were attributed to soil variations and the release of potash by lime. However, Abel and Magistad (*1*) found that more potassium was removed by crops from the limed soil than from the unlimed. Analysis of hay from a few plots indicated that more potash was removed from the limed plots. Unfortunately, a sufficient number of analyses was not made nor were the yield data sufficiently differentiated to permit a calculation of the total potassium removed by the crops during the period involved. However, the results indicate the possibility that the losses by leaching may have been sufficiently lower in the limed soils to account for the higher total potassium content. This is supported by the results obtained by Abel and Magistad (*1*), who found that with a soil deficient in available potash the losses by leaching were greatly reduced while the removal by crops was increased by liming. The soil employed in the present investigation is low in exchange capacity and potash is the first limiting factor in crop growth.

In view of the results of recent investigations and the evidence obtained under the conditions outlined, it appears that there is a retention of potassium brought about by liming and that at least a part of the potassium is held in a form which, while available to plants, is incapable of entering the soil solution. This potassium is undoubtedly more available to plants capable of utilizing larger amounts of calcium, for as the calcium is removed by the plant the equilibrium is shifted in the direction in which potassium ions enter the system. This action takes place at the root-soil interface, and since the potassium is immediately utilized by the plant, the free soil solution is not enriched in potassium ions. Consequently, the losses of potassium by leaching are less than in the unlimed soils. The greater exchangeable and electrodialyzable potassium content of the limed soils may be accounted for in part by an increase in exchange capacity brought about by larger crop residues and higher pH.

SUMMARY

Fertilizer plot soils were subjected to electrodialysis and leaching with neutral ammonium acetate to determine the effect of fertilizers, cropping, and liming upon the amount of potassium removed by these methods.

Without exception, more potash was removed by electrodialysis from the limed soils than from the unlimed. The results for exchangeable potash were similar but of lesser magnitude.

Applications of potash were reflected to a greater degree by electrodialysis than by the exchange method.

Exchangeable and dialyzable potash were at a maximum in the plot receiving manure plus lime.

pH values were remarkably uniform throughout each series (limed and unlimed) of soils.

Without exception, total potash content of the soil was greater in the limed series.

For the soil investigated it appears that liming has conserved potash by reducing losses from leaching.

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AN EXPERIMENTAL STUDY OF THE RELATION BETWEEN THE UROPYGIAL GLAND AND VITAMIN D DEFICIENCY IN CHICKS [1]

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INTRODUCTION

The part played by the uropygial gland in the physiology of birds has been the subject of speculation for many years. Recent discoveries relating to vitamin D and the role of ultraviolet radiation in its elaboration have stimulated interest in the gland.

In 1913 Paris (*12*)[3] published the results of a very thorough experimental investigation of the structure and function of the uropygial gland. He, and others to whom he refers, concluded that the gland serves no indispensable purpose, for upon occlusion or extirpation of the gland no differences were observed between the experimental birds and the normal controls.

Rowan (*13*) noted that the preen gland is rich in cholesterol, and suggested that feather or feather oil may be a source of vitamin D.

Hou (*4*, *5*, *6*) investigated the relation of the uropygial gland to the production of vitamin D. His experimental evidence tended to show that the uropygial gland is the sole source of vitamin D precursor available to the bird. The results of further investigation (*7*) apparently substantiated this view. In a later publication, however, (*8*) he admitted that some other substance might be involved in vitamin D elaboration.

The fact that many birds lack the gland is not concordant with the idea that it performs a definite and necessary function.

The present investigation was undertaken with the relation between the uropygial gland and vitamin D production particularly in mind.

EXPERIMENTAL PROCEDURE

In the winter of 1930–31 and again in the fall of 1931, experiments were made to determine whether or not the removal of the uropygial gland from chickens would prevent the utilization of ultraviolet radiation in the production of vitamin D. The results of these two experiments indicated that the gland is dispensable in the calcium metabolism of the bird. The irradiated birds without the glands grew as well as the normal irradiated birds, and the ash content of their bones was as high.

On December 6, 1933, a third experiment was begun, which was carried out under carefully controlled conditions. One hundred Rhode Island Red chicks of medium weight were selected, and from 50 of these, the uropygial glands were extirpated. The combs were

[1] Received for publication July 11, 1935; issued August, 1936. Paper 210 of the biological laboratory of the Maine Agricultural Experiment Station. A report of this investigation in thesis form was filed in the library of the University of Maine in June 1934.

[2] The writer is indebted to Dr. W. F. Dove for suggesting the problem and for help and advice in carrying on the research, and to Mildred R. Covell for preparing the graphs.

[3] Reference is made by number (italic) to Literature Cited, p. 71.

removed from all of the birds to preclude the possibility of comb development interfering with the results (Maughan and Dye (*11*)).

Until the birds were put on experiment at 3 weeks of age, all of them were fed a mash ration containing 2 percent of cod-liver oil. This preliminary treatment is in accordance with the recommendation of Dove (*2, pp. 219–223*). The glands were removed when the birds were 2 weeks of age, and 1 week was allowed for recovery from the operation before the experimental treatment was begun. The birds were fed Hart's rachitogenic ration (*3*). This, the basic ration, was made up according to the following formula:

59 parts ground yellow corn.
25 parts standard wheat middlings.
12 parts crude casein.
1 part common salt.
1 part precipitated calcium carbonate.
1 part precipitated calcium phosphate.
1 part dried yeast.

Some of the birds were subjected to ultraviolet prophylactic treatment and others to ultraviolet curative treatment. The prophylactic groups received 5 minutes daily ultraviolet irradiation instead of a fully protective dose of 20 minutes. If the presence of the uropygial gland were essential for normal calcification, the relation would be apparent at this lower level of protection.

Some control groups of birds received the rachitogenic ration supplemented with cod-liver oil. Other control groups were fed the basic ration without any vitamin supplement. Groups of chickens with the glands intact and groups with the glands removed were kept under similar conditions and subjected to similar treatments.

EXPERIMENTAL RESULTS

The results of the experimental treatments are shown in table 1. Each of the 10 groups consisted of 10 birds, some male and some female. Each bird was weighed at weekly intervals. The weekly weights are presented in figure 1, *A*, for the groups receiving prophylactic treatment, and in figure 1, *B*, for the groups receiving curative treatment.

TABLE 1.—*Treatment of chickens in the various groups of 10 birds each, and percentage ash of fat-free bones*

Group no.	Treatment [1]	Average percentage of ash based on fat-free bone
295	Gland intact; curative treatment; basic ration plus ½ percent Mazoil for 5 weeks, then supplemented with 20 minutes daily ultraviolet radiation for remaining 4 weeks of experiment	41.61
300	Gland removed; curative treatment; basic ration plus ½ percent Mazoil for 5 weeks, then supplemented with 20 minutes daily ultraviolet radiation for remaining 4 weeks of experiment	42.35
296	Gland intact; curative treatment; basic ration plus ½ percent Mazoil for 5 weeks, then ½ percent cod-liver oil substituted for Mazoil.[2]	44.70
301	Gland removed; curative treatment; basic ration plus ½ percent Mazoil for 5 weeks, then ½ percent cod-liver oil substituted for Mazoil	43.82
293	Gland intact; control; basic ration plus ½ percent Mazoil	36.37
298	Gland removed; control; basic ration plus ½ percent Mazoil	38.14
294	Gland intact; preventive treatment; basic ration plus ½ percent Mazoil plus 5 minutes daily ultraviolet radiation	40.73
299	Gland removed; preventive treatment; basic ration plus ½ percent Mazoil plus 5 minutes daily ultraviolet radiation	40.71
297	Gland intact; preventive treatment; basic ration plus ½ percent cod-liver oil	49.17
302	Gland removed; preventive treatment; basic ration plus ½ percent cod-liver oil	47.75

[1] The basic ration was Hart's rachitogenic ration; Mazoil is a commercial corn oil.
[2] 4 of the birds died in the fourth week of the experiment from accidental causes.

In the prophylactic test, the irradiated birds with the glands removed gained less than the irradiated birds with the glands intact. These differences in growth, however, were too small to justify an assumption that the presence or absence of the gland was directly responsible for them. In the curative test, the birds from which

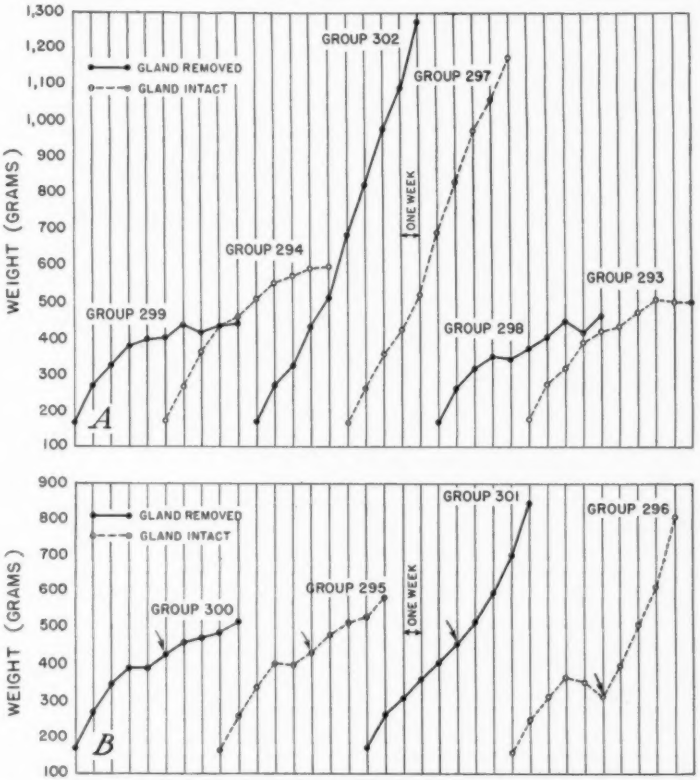

FIGURE 1.—Average weekly weight of chicks on preventive (*A*) and on curative (*B*) tests. Arrows in *B* indicate points at which daily irradiation with ultraviolet light was initiated for groups 295 and 300, and at which the cod-liver oil supplement was added to the basic (Hart's rachitogenic) ration consumed by groups 296 and 301. Groups 293 and 298 (controls) were on the unsupplemented rachitogenic ration; this was supplemented, as explained in the text, with ultraviolet ray irradiations in groups 294, 295, 299, and 300, and with cod-liver oil in groups 296, 297, 301, and 302. The uropygial gland was removed from all birds in groups 298 to 302 and was intact in groups 293 to 297.

the glands had been removed and which were subsequently allowed to become rachitic showed a definite improvement in growth upon the initiation of daily irradiation.

No differences in the calcification of the bones were apparent between comparable irradiated groups. The ash content of the bones of the birds without the glands was as high as that of the bones of the birds with the glands.

DISCUSSION

It is difficult to reconcile the data here reported with the results of Hou's investigations. His experimental evidence tends to establish his premise that birds from which the uropygial glands are extirpated are unable to utilize ultraviolet radiation in the production of vitamin D. He concluded that the bird is dependent upon the uropygial secretion for vitamin D precursor.

The writer's experiments fail to substantiate this hypothesis. Since these experiments were completed, Knowles, Hart, Halpin, and Holmes, according to Clark (*1*), have completely refuted the conclusions of Hou. They were unable to discover any relation between the uropygial gland and the elaboration of vitamin D. In every case they found that the birds from which the glands had been removed grew as well as those in which the glands were intact. Knowles, Hart, and Halpin (*9*) published their data in detail, including bone analyses, which showed normal calcification in birds both with and without the gland. The writer's results, therefore, coincide with the conclusions of their investigations.

The origin of activatable ergosterol in the bird is still an open question. Maughan (*10*) implies that activation in chickens occurs in the circulating blood. Maughan and Dye (*11*) found that female chickens require double the irradiation required by males. They attributed this fact to the greater comb development of the males and the consequent larger area of uncovered body surface exposed to the rays. In the same paper Maughan and Dye report that exposure of the legs alone to ultraviolet radiation causes as rapid a recovery from rickets as total irradiation. Hou (*8*) stated his belief that the skin of the legs is too thick and the circulation too scanty to give credence to the hypothesis that the circulating blood contains the activatable ergosterol.

As far as the production of vitamin D is concerned the uropygial gland possibly augments the supply of ergosterol available in sufficient amounts from some other source. From the data presented the writer feels that it is safe to assume that the uropygial gland serves no indispensable function.

SUMMARY AND CONCLUSION

In a carefully controlled experiment the uropygial glands were extirpated from chicks in order to discover any relation that might exist between the presence of the gland and the ability of the birds to utilize ultraviolet radiation in the production of vitamin D.

Groups of birds with the glands removed and groups of birds with the glands intact were treated with ultraviolet radiation. Some groups were subjected to prophylactic and some to curative treatment. Control groups of birds, with and without glands, were fed the basic rachitogenic ration supplemented with Mazoil. Other control groups were given the basic ration supplemented with cod-liver oil.

The average weekly weights of the preventive groups, presented in graphic form, show small differences between the comparable groups receiving daily radiation.

It is evident from the graphic representation of average weekly weights that rachitic birds from which the uropygial glands had been removed definitely improved upon daily irradiation.

The bone analyses showed no differences in total ash content between groups with the glands removed and those with the glands intact.

It is concluded that, in chicks, the lack of the uropygial gland does not preclude the utilization of ultraviolet radiation in the production of vitamin D.

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OXIDATION-REDUCTION POTENTIALS OF SOIL SUSPENSIONS IN RELATION TO ACIDITY AND NITRIFICATION[1]

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INTRODUCTION

Since Gillespie, (*2*),[2] in 1920, first measured the potential in a soil, using an indifferent electrode, numerous investigations have been made concerning the methods of measuring this oxidation-reduction potential. Several soil properties have been shown to be empirically associated with the potential, and numerous soil phenomena have been claimed, but not demonstrated to result from high or low potentials. For the most part, these investigations have been concerned with the methods of determination and the effects of artificial changes on the value of the potentials. There is a need for more fundamental study of the cause-effect relationships of the oxidation-reduction potential to other soil properties.

OBJECT OF EXPERIMENT

Of all the oxidation-reduction systems in the soil the most important, from an agronomic point of view, are those of carbon, nitrogen, and oxygen. The relation of the reversible oxygen system to the oxidation-reduction potential has already been established (*8*). That the carbon system is reversible has not been demonstrated, but Latimer and Hildebrand (*6*), in a table of potentials, indicate that the ammonia-nitrate system is reversible and has a definite potential at given concentrations. The statement of Heintze (*4*) that "in standardized media the form of the potential-time curve can serve to characterize groups of microorganisms or cellular processes, e. g., dentrification" leaves the conclusion to be drawn that the nitrogen system, when present in great enough concentration, should play a major part in establishing the potential of a soil.

A review of the literature has revealed only one study of soil oxidation-reduction potentials as they vary over a period of weeks; and that one (*7*) is an empirical study of potentials rather than an analysis of simultaneous changes which might cause the changes of potential. The objects of this experiment, therefore, have been (1) to study the changes in the oxidation-reduction potentials of suspensions of different soils over periods of time long enough to permit biological activity to cause significant changes in the nitrogen system, and thereby to ascertain whether the nitrogen system plays a part in establishing the potentials of soils; and (*2*) to note any changes in the oxidation-reduction potentials which may be ascribed to soil acidity or fertilizer treatment.

[1] Received for publication June 3, 1935, issued August, 1936.
[2] Reference is made by number (italic) to Literature Cited, p. 80.

PROCEDURE

Various soils were limed at abnormally high rates, and different form of nitrogen were applied. Periodic analyses of nitric and ammonic nitrogen, acidity, and oxidation-reduction potential were made over an 8-weeks' course of nitrification in the greenhouse.

The soils used were: (1) A well-drained sandy loam pasture soil; (2) the same soil in a natural waterlogged condition, showing the growth of sedges; (3) a gravelly sand, taken from a railroad fill; and (4) a black, sandy soil taken from a swamp.

About 400 pounds of each moist soil was collected, and after the soils had dried for 2 to 3 weeks, the clods were broken up and the samples were sifted twice through a three-mesh sieve, mixed several times, and divided into two parts. To one lime was added at the rate of 7½ tons per acre (58.5 percent Ca $(OH)_2$, 41.5 percent $CaCO_3$); to the other no lime was added. One week later, both the limed and the unlimed portions were again subdivided into four 8-kg parts each. Nitrogen was then applied as explained in table 1, after which the 32 resulting samples were placed, to a depth of about 4 inches, in wooden boxes 1 foot square by 6 inches deep, so constructed as to leave a quarter-inch opening along the bottom, which was covered with a paper towel. The boxes were placed on a greenhouse bench and watered at least once a week; the moisture content was kept between 3 and 25 percent.

Samples were taken at the time of nitrogen application and at intervals of approximately 2 weeks, for the determination of moisture, nitrate nitrogen, ammonia nitrogen, acidity, and oxidation-reduction potential.

Determinations of pH were made with the quinhydrone electrode. About 15 g of air-dry soil, 0.05 g of quinhydrone, and a 1: 2 soil-water ratio were used.

TABLE 1.—*Description and treatment of soils*

Soil no.	Description	Lime applied	Nitrogen applied
100	Well drained, fine sandy loam, from Brooks' farm, Massachusetts State College.	None	None.
101		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
102		do	1 g dried blood per 100 g soil (10 percent N).
103		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
110		7½ tons per acre	None.
111		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
112		do	1 g dried blood per 100 g soil (10 percent N).
113		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
200	Poorly drained, fine sandy loam, from Brooks' farm, Massachusetts State College.	None	None.
201		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
202		do	1 g dried blood per 100 g soil (10 percent N).
203		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
210		7½ tons per acre	None.
211		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
212		do	1 g dried blood per 100 g soil (10 percent N).
213		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
300	Gravelly sand fill, from Tillson farm, Massachusetts State College.	None	None.
301		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
302		do	1 g dried blood per 100 g soil.
303		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
310		7½ tons per acre	None.
311		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
312		do	1 g dried blood per 100 g soil.
313		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
400	Black, sandy swamp soil, from Tillson farm, Massachusetts State College.	None	None.
401		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
402		do	1 g dried blood per 100 g soil.
403		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.
410		7½ tons per acre	None.
411		do	30 mg NH_3–N per 100 g soil, as $(NH_4)_2SO_4$.
412		do	1 g dried blood per 100 g soil.
413		do	30 mg NO_3–N per 100 g soil, as $NaNO_3$.

Nitrate nitrogen determinations were made colorimetrically by the phenoldisulphonic method. Ammonia nitrogen determinations were made according to Harper's method (*3*).

The oxidation-reduction potentials were determined by a modification of Willis' method (*8*). The modification consisted chiefly in the omission of the displacement of air with nitrogen, which Herzner (*5*), Brown (*1*), and Heintze (*4*) have shown to be unnecessary. An experiment conducted by the senior writer in which the two methods were compared showed that the results by the modified method can be rather well duplicated on different samples of the same soil, and that the displacement of air is not necessary.

Fifty grams of moist soil were placed in a 250-cc flask with an equal weight of distilled water. The flask was stoppered and shaken at room temperature (about 28° C.) for 2½ to 3 days. The contents of the flask were then transferred to a 250-cc three-necked Woulff bottle, two bright platinum electrodes were inserted and the bottle was placed in a constant-temperature water bath at 35° C. Connection to a saturated calomel half-cell was made with a 3-percent agar-saturated potassium chloride bridge. Within 15 minutes of contact of the electrodes with the soil suspension, the potential of the resultant cell,

Hg | HgCl (sat.), KCl ‖ Agar–KCl ‖ soil suspension | Pt,

was measured electrometrically on a Leeds & Northrop portable acidity meter. The duplicate electrodes always checked within 5 millivolts and generally within a narrower range.

Between determinations on different soils, the agar bridge was rinsed with saturated KCl; between weekly runs, it was set in saturated KCl to prevent desiccation. Before and after each determination on different soils, the platinum electrodes were rinsed with water, then with alcoholic alkali, alcohol, and water. Between weekly runs, the electrodes were set in distilled water.

RESULTS

The results of the experiment are given in tables 2 and 3. The results from six representative soils are shown graphically in figure 1. Nitric and ammonic nitrogen were expressed as parts of nitrogen per 10,000 of moisture-free soil. Acidity was expressed in pH values. Since the values of soil potentials found by the majority of previous investigators seem to indicate that they have been expressed as the potentials of the half-cell in tenths of volts,

Soil suspension ‖ Pt,

the potentials observed in this experiment have been expressed in the same manner, for purposes of comparison. Although this is not in accord with the convention followed in most textbooks, viz, the expression of such potentials as those of the cell above, reversed, there is no essential difference as long as it is specified which half-cell is being spoken of, for in either case the interpretation is that the oxidation-reduction system of the soil is more oxidative than the standard hydrogen electrode.

TABLE 2.—*Concentrations of ammonic nitrogen and of nitric nitrogen in soils* [1] *on different dates*

Soil no.	Feb. 7		Feb. 23		Mar. 10		Mar. 20		Apr. 4	
	NO_3–N	NH_3–N	NO_3–N	NH_3–N	NO_3–N	NH_3–N	NO_3–N	NH_3–N	NO_3–N	NH_3–N
	P. p. m.	*P. p. m.*	*P. p. m.*	*P. p. m.*	*P. p. m.*	*P. p. m.*	*P. p. m.*	*P. p. m.*	*P. p. m.*	*P. p. m.*
100	15	38	23	30	52	24	67	14	62	4
101	15	344	16	340	34	292	26	308	103	90
102	66	72	62	431	106	432	111	418	182	556
103	300	37	163	38	444	50	361	52	487	46
110	17	36	47	30	71	17	44	29	89	7
111	16	283	220	47	353	23	313	72	370	37
112	13	61	177	377	236	307	390	195	589	207
113	275	31	483	44	487	38	265	5	470	7
200	48	31	79	49	80	51	85	43	82	33
201	32	326	32	310	69	406	21	331	88	369
202	43	60	82	------	208	288	246	417	285	484
203	278	31	353	56	462	67	306	63	461	74
210	46	31	51	47	164	23	113	6	175	3
211	26	278	193	79	523	47	388	9	382	3
212	47	58	186	351	551	121	471	22	881	43
213	321	30	326	70	259	40	382	31	500	28
Soil no.	Feb. 15		Feb. 28		Mar. 16		Mar. 28		Apr. 10	
300	10	14	14	75	23	7	36	8	49	6
301	10	334	15	340	23	236	41	194	63	247
302	10	55	27	425	68	432	122	357	172	444
303	360	18	228	66	324	32	315	27	324	37
310	10	21	27	72	116	20	135	23	201	5
311	9	321	20	235	192	135	263	78	520	40
312	10	70	23	463	61	349	52	325	240	363
313	312	21	282	105	340	63	471	56	545	41
400	6	34	5	82	14	64	22	64	73	36
401	3	258	Trace	283	Trace	360	Trace	379	Trace	477
402	3	82	12	595	15	686	11	689	53	783
403	313	31	294	79	316	87	396	96	358	114
410	3	46	20	85	97	31	129	10	108	28
411	3	333	19	357	148	161	271	111	141	76
412	4	98	42	552	595	157	321	178	807	9
413	307	41	220	87	385	58	554	21	328	10

[1] See table 1 for soil treatments; all determinations calculated on the basis of moisture-free soil.

TABLE 3.—*Acidity and oxidation-reduction potentials* [1] *of soil* [2] *suspensions on different dates*

Soil no.	Feb. 11		Feb. 24		Mar. 11		Mar. 20		Apr. 4	
	pH	E_h	pH	E_h	pH	E_h	pH	E_h	pH	E_h
		Volt		*Volt*		*Volt*		*Volt*		*Volt*
100	5.47	+0.488	5.38	+0.532	5.21	+0.520	4.90	+0.571	5.19	+0.600
101	5.46	+.493	5.31	+.532	5.21	+.530	4.83	+.603	4.89	+.615
102	5.88	+.486	5.79	+.488	5.64	+.500	5.44	+.452	5.40	+.499
103	5.43	+.484	5.38	+.571	5.17	+.544	5.01	+.583	5.14	+.585
110	7.31	+.386	7.26	+.435	6.47	+.448	6.56	+.434	5.98	+.486
111	7.74	+.386	7.10	+.431	6.73	+.447	6.97	+.441	6.09	+.485
112	7.84	+.040	7.70	+.343	7.38	+.365	7.11	+.419	6.61	+.467
113	7.62	+.387	7.65	+.433	7.46	+.441	7.44	+.442	7.38	+.487
200	5.48	+.479	5.36	+.560	5.36	+.558	5.07	+.576	5.25	+.575
201	5.42	+.479	5.35	+.561	5.34	+.556	4.98	+.589	5.05	+.589
202	5.87	+.462	5.95	+.494	5.29	+.556	5.02	+.569	5.19	+.583
203	5.43	+.477	5.33	+.560	5.22	+.563	5.01	+.582	5.14	+.583
210	7.30	+.357	7.55	+.448	6.93	+.463	7.24	+.466	7.24	+.473
211	7.48	+.362	7.31	+.454	6.90	+.474	7.09	+.489	7.31	+.501
212	7.91	+.085	7.50	+.358	7.05	+.450	7.03	+.490	6.98	+.507
213	7.64	+.356	7.75	+.442	7.53	+.449	7.62	+.481	7.65	+.487
Soil no.	Feb. 15		Mar. 2		Mar. 16		Mar. 28		Apr. 10	
300	5.28	+0.523	5.04	+0.558	4.65	+0.553	4.47	+0.581	4.85	+0.614
301	5.11	+.524	4.89	+.575	4.51	+.575	4.27	+.597	4.60	+.617
302	5.32	+.471	5.47	+.093	5.80	+.343	4.98	+.457	5.00	+.403
303	5.06	+.560	4.87	+.575	4.63	+.568	4.49	+.568	4.84	+.533
310	8.07	+.328	7.46	+.389	6.87	+.392	6.70	+.435	7.00	+.513
311	8.16	+.321	7.74	+.388	7.23	+.374	6.90	+.426	6.30	+.503
312	8.17	+.020	8.07	+.030	7.42	+.030	7.35	+.038	7.52	+.377
313	8.09	+.318	7.80	+.400	7.55	+.382	7.03	+.415	7.54	+.458
400	5.15	+.491	5.30	+.537	4.82	+.540	4.74	+.556	5.02	+.590
401	5.16	+.499	4.98	+.544	4.71	+.564	4.68	+.535	5.00	+.570
402	5.39	+.391	5.52	+.265	5.50	+.306	5.42	+.368	5.49	+.477
403	5.16	+.504	4.95	+.535	4.81	+.536	4.70	+.543	5.00	+.560
410	7.01	+.378	6.47	+.429	6.00	+.447	6.18	+.476	6.08	+.496
411	7.64	+.372	6.91	+.440	6.55	+.458	6.15	+.460	6.21	+.500
412	7.68	+.103	7.33	+.191	6.53	+.355	6.32	+.403	6.10	+.517
413	7.64	+.387	7.05	+.413	7.06	+.419	6.67	+.479	6.71	+.499

[1] Expressed as the potential of the half-cell, | soil suspension | Pt, at 35° C.
[2] See table 1 for soil treatments.

Nitrification, changing acidity, and varied nitrogen treatments were considered as possible causes of changes of potential. For comparing nitrification and potential, two methods were used: (1) The increase of nitrate nitrogen over the original determination, the decrease of ammonia nitrogen, and the sum of these two values, considered as

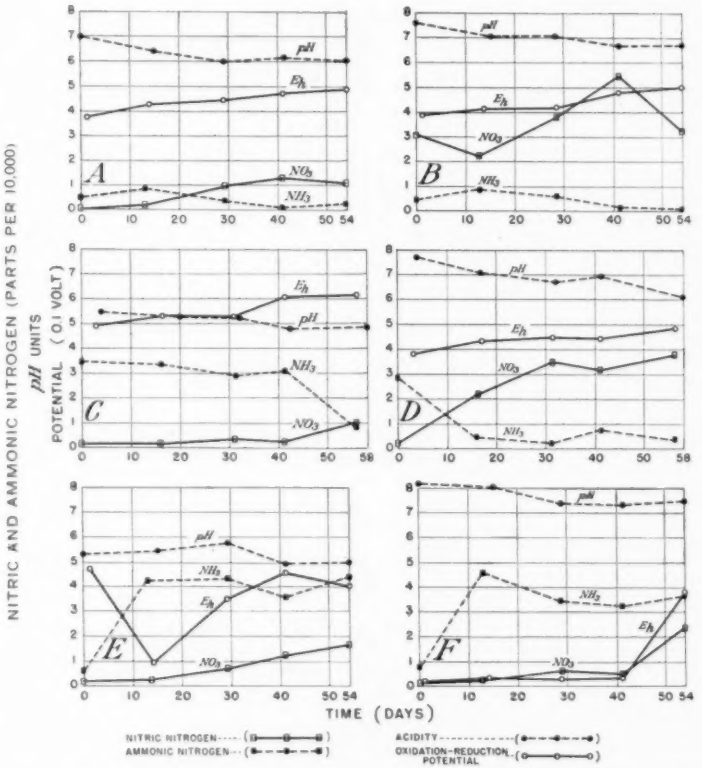

FIGURE 1.—Nitric and ammonic nitrogen in parts per 10,000 of moisture-free soil, acidity as pH, and half-cell potential, of six soils during the experimental period: *A*, Soil 410, poorly drained, treated with lime, no nitrogen, in tenths of volts; *B*, soil 413, poorly drained, treated with lime and $NaNO_3$; *C*, soil 101, well-drained, treated with $(NH_4)_2SO_4$, no lime; *D*, soil 111, well-drained, treated with lime and $(NH_4)_2SO_4$; *E*, soil 302, well-drained, treated with dried blood, no lime; *F*, soil 312, well-drained, treated with lime and dried blood.

giving a measure of the amount of nitrification, were compared with the simultaneous increase of potential over its initial value; (2) values of log $(NO_3\text{–}N)/(NH_3\text{–}N)$ were calculated at each determination, and plotted on scatter diagrams against simultaneous values of potential. In comparing potentials with acidity, the coefficients of correlation and regression were calculated as explained below. In the comparison of potential with nitrogen treatment, the initial potentials of nitrogen-treated soils were compared with the potentials of their corresponding no-nitrogen treatment.

CHANGES OF POTENTIAL WITH RELATION TO NITRIFICATION

In general, since nitrification took place at the expense of the ammonia content of the soils, the NO_3–N and NH_3–N concentrations showed a rough inverse trend, except in those soils where there was considerable ammonification of added or already present organic matter.

The increases of nitrate content over the initial determination for each soil were calculated at 2, 4, 6, and 8 weeks. In like manner, the decreases of ammonia content and the values of (increase nitrate+decrease ammonia) were calculated. These measures of nitrification were compared with the increases of potentials observed at corresponding times.

This method of comparison showed no definite relationship within the individual soils. In nearly all soils the potentials showed steady increases. But as the potential of a soil increased, the nitrification often decreased. For example, in soil 203 the increases in potential over the initial value were 83, 86, 105, and 106 mv, while the corresponding nitrate increases were 75, 184, 28, and 183 p. p. m., the ammonia decreases were −25, −36, −32, and −43 p. p. m., and the values of (increase NO_3+decrease NH_3) were +50, +148, −4, and +140 p. p. m.

When the increase for any limed soil was compared with the simultaneous increase for its corresponding unlimed soil, however, there appeared to be a more definite relationship. In all but 9 of the 64 simultaneous comparisons of increase of potential, the increase was more positive in the limed soil than in the unlimed. Likewise, in all but 10 and 12 comparisons, respectively, the increase of nitrate and the decrease of ammonia were more positive in the limed soil than in the corresponding unlimed soil. This relationship was merely qualitative, as the magnitudes of increases of potential were not proportional to increases of nitrate.

The final criterion, however, of the effect of any oxidation process on potentials must be the variance of potential with changing ratios of oxidant to reductant. Bearing this in mind, and the equation for the potential of any reversible oxidation-reduction system

$$E_h = E_o + \frac{RT}{nF} \log \frac{(\text{oxidant})}{(\text{reductant})},$$

values of $\log \left(\frac{NO_3-N}{NH_3-N} \right)$ were calculated for each simultaneous determination. With these values as abscissae and the corresponding observed values of E_h as ordinates, scatter diagrams were constructed for various combinations of soils, as follows: (1) All soils; (2) all unlimed soils; (3) all limed soils; (4) all limed soils except those receiving dried-blood treatment; (5) all different lime and nitrogen treatments of each of the four original soils; (6) each similar nitrogen treatment of all soils.

The only combination in which the points appeared to be grouped along a straight line was combination 4, of all limed soils except the dried-blood-treated. In this combination there appeared to be a slight positive correlation of E_h to log $(NO_3–N)/(NH_3–N)$. The points for the dried blood treatments, however, after the first one or two determinations, also fell fairly well within the same range. This strengthened the proposition that oxygen depletion was effec-

tive in lowering the potentials during the initial stages, when decomposition of organic matter and ammonification were rapid.

To summarize: Changes in potential were more closely correlated with changes in acidity than with changes in the nitrogenous constituents. The addition of large amounts of nitrate nitrogen or ammonia nitrogen had very little effect on the potentials of suspensions of the soils. The apparent correlation of potentials with $\log(NO_3/NH_3)$ held only in limed soils, where nitrification was very active and where there were large changes in acidity.

As a general rule, the process of ammonification is considered as an alkalizing process, and that of nitrification as an acidifying process. The results of this experiment were interpreted as being in accord with this relationship, viz, when the microbiological processes of ammonification and nitrification were active in the soils, a change was reflected in the potentials observed on suspensions of the soils. The changes in potential were more closely correlated with changes in acidity than with changes in the nitrogenous constituents, suggesting that the resultant acidity or alkalinity, rather than the presence of nitrogen as ammonia or nitrate, was the cause of changes in potential.

CHANGES OF POTENTIAL WITH RELATION TO CHANGING ACIDITY

The observations of Herzner (*5*), Willis (*8*), and Heintze (*4*), that the potential is a direct function of acidity were remarkably well verified by the results of this experiment. This relationship held equally well for changes of acidity produced by liming and for those produced in the course of time along with changes in the nitrogenous constituents.

Eliminating the dried blood treatments, in which the pH–E_h relationship seemed to be complicated by the rapid decomposition of organic matter, the remainder of the simultaneous pH and E_h readings showed a decidedly negative correlation. The correlation coefficient of these variables at 0, 2, 4, 6, and 8 weeks was, respectively, -0.97 ± 0.0081, -0.96 ± 0.0108, -0.92 ± 0.0211, -0.74 ± 0.0623, and -0.90 ± 0.0262. When combined without respect to time, all the pH and E_h readings except those of the dried-blood treatments showed a gross correlation coefficient of -0.85 ± 0.0171. The regression coefficient, which is a measure of the increase of potential with a unit decrease of pH, was, at the same intervals, 0.0594, 0.0575, 0.0553, 0.0381, and 0.0479. The regression coefficient of all the readings except those of the dried blood treatments, combined without respect to time, was 0.0540. These figures compare rather favorably with the figure 0.0611, which is the theoretical regression coefficient of the potential of a monoelectronic oxidation-reduction system with pH, at 35° C. Although this may not be conclusive, since the homogeneity and size of the population have not been statistically validated, it seems to indicate that the hydrogen-hydrogen ion system is very important in determining the magnitude of the oxidation-reduction potentials of soils.

CHANGES OF POTENTIAL DUE TO APPLICATION OF NITROGEN CARRIERS

The potentials of suspensions of different nitrogen treatments of the same soil, when compared with that of the no-nitrogen treatment, showed only slight, inconsistent changes in the cases of the $NaNO_3$ and the $(NH_4)_2SO_4$ treatments. This lack of change confirmed the idea stated above that changes in ammonia and nitrate content effect

changes of potential only as these changes alter the acidity of the soil. The treatments with dried blood, on the other hand, resulted in marked decreases of potential, particularly at high pH values. In these soils there was a rapid decomposition of the added organic matter, as was indicated by the large rises in ammonia content during the first 2 weeks of the experiment. During this period, the potentials of suspensions of the dried-blood-treated soils were from 50 to 350 mv lower than would be expected from the pH–E_h relationship which has been demonstrated for the other soils. It has been shown by Willis [3] that oxygen depletion should cause a drop in potential if the pH remains the same. It is suggested that in such cases as these dried-blood-treated soils, the rapid decomposition of organic matter was accompanied by oxygen depletion, which caused the drops in potential.

SUMMARY

By means of a greenhouse study of four soils from different drainage conditions, at different lime levels, and with different nitrogen fertilization, the relationships of the oxidation-reduction potentials of suspensions of these soils to nitrification, acidity, and kind of nitrogen carrier were investigated.

It was found that any change of potential with nitrification is secondary to the effect of nitrification on pH.

The inverse relationship found independently by Herzner, Willis, and Heintze to exist between pH and E_h was verified.

The addition to the soils of $(NH_4)_2SO_4$ or $NaNO_3$ was found to have no constant effect on the potentials of the suspensions.

The rapid decomposition of organic matter brought about a marked fall of potential. This fall was apparently caused by oxygen depletion.

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U. S. GOVERNMENT PRINTING OFFICE: 1936